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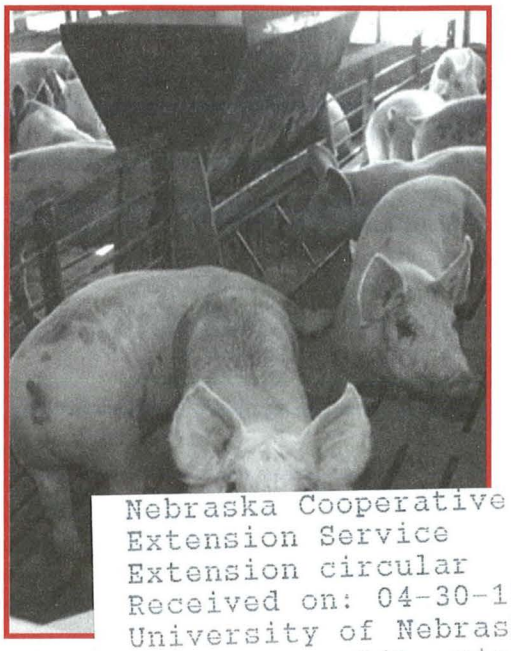

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Prepared by the staff in Animal Science Department and cooperating departments for use in Extension, Teaching, and Research programs.

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Table of Contents

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2010 Nebraska Swine Report

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Production Through Four Parities of Prolific Females Developed With and Without Energy Restriction

A 25% energy restriction during development delays sexual development of gilts but has no effect on the reproductive rate of those reaching sexual maturity.

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Summary

This experiment evaluated the effects of developing gilts with ad libitum access to feed to breeding age (226 days) or feed intake restriction from 123 to 226 days of age. Gilts were managed in groups of 10 per pen. Those in the restricted group were fed two meals per day so that energy intake was 75% of that of the ad libitum group. Protein, vitamins, and minerals in their diet were increased so that daily intake of these nutrients was not restricted. A total of 661 gilts of two genetic lines that differed in reproductive rate and in lean growth rate started the experiment at 60 days of age, and one-half of the gilts of each line were developed with each feeding regimen. Growth and backfat were recorded at 14-day intervals from 60 to 226 days of age. Boar exposure to determine age at puberty was initiated at 140 days of age. A total of 509 gilts that could be mated at second or later post-pubertal estrus were designated as breeders and their production through four parities was recorded. Females were managed alike after 230 days of age and were culled only for reproductive failure, death, ruptures, or severe foot and leg problems. No interactions of genetic line by treatment were significant as females of both lines responded similarly to the developmental regimens. Developing gilts with energy restriction significantly decreased the proportion of gilts that expressed a pubertal estrus by 230 days of age, from 96% to 86% and increased their age at puberty from 174.1 to 177.5 days. Thereafter, females developed with both regimens had similar reproductive performance. Measures of productivity through parity 4 were 8 to 11% greater for females developed with energy restriction, but none of the differences were significant ($P \geq 0.14$).

Introduction

In several species, restricting energy intake postweaning, without limiting other nutrients, often increases longevity. Sometimes, reallocation of resources occurs such that animals cannot combine high rates of fecundity with extended lifespans. However, this outcome does not always happen. In one study, mice restricted in energy intake postweaning lived longer without a reduction in reproductive rate (Johnston et al., 2006, Proc. R. Soc. B 273:1369-1374). Consistent with these results, a series of experiments at the USDA Meat Animal Research Center (Klindt et al, 1999, J. Anim. Sci. 77:1968-1976, 2001, 79:787-795, and 2001, 79:2513-2523) demonstrated that moderate feed restriction during prepubertal development of gilts may increase reproductive efficiency through first parity.

Today's commercial gilts are often managed to achieve weights of at least 136 kg (300 lb) before they are mated or inseminated, and it is generally believed that some minimum amount of backfat is needed for reproduction. Thus, gilts are often developed to breeding age with ad libitum access to feed. The experiments cited above indicate that this management strategy may be detrimental to long-term reproductive performance. Therefore, we designed an experiment to examine whether restricting energy intake during a gilt's developmental period will increase their longevity and lifetime productivity.

Because optimum gilt development regimens may vary among genetic lines, depending on the line's prolificacy and rate of lean growth, gilts of two lines that differed in fertility, litter size, and rate of lean growth were managed with ad libitum access to feed to breeding age or with 25% restriction of energy from 123 days of age to breeding. Reproduction through parity 4 was evaluated. The experiment was done in four replications with a total of 661 gilts. Reproductive performance of females through parity 4 for gilts of replications 1 to 3 are in the *2008 Nebraska Swine Report* (Miller et al., 2008; Johnson et al., 2008). A fourth replication was subsequently added to the project, and the *2009 Nebraska Swine Report* summarizes productivity of all females through parity 1. With completion of fourth parity litters by replication 4 females in summer 2009, the experiment is complete, and production of all females through 4 parities is summarized here.

Materials and Methods

Gilt Populations

Two populations of gilts were used. One was the Large White by Landrace crossbred female used in the UNL swine nutrition program. The project gilts

(Continued on next page)

were the progeny of Large White-Landrace cross sows that had been inseminated with semen of industry maternal line (L_M) boars and are designated as LW x LR cross. The other population, Line 45X, were progeny of UNL selection Line 45 sows that had been inseminated with semen of the same L_M boars used to produce LW x LR gilts. Line 45 has now undergone 29 generations of selection for increased litter size with additional selection for increased growth and decreased backfat in the last seven generations. The Line 45 dams used to produce gilts for the experiment were from generations 25, 26, and 27. Based on previous data, L45X gilts are expected to be more prolific than LW x LR gilts, but also have somewhat slower growth and greater backfat thickness.

Gilt Management and Dietary Regimens

Project gilts were born in batches during December 2004 and January 2005 (Rep 1), May 2005 (Rep 2), November 2005 (Rep 3), and May and June 2007 (Rep 4). A total of 661 gilts began the experiment (157 to 185 gilts per replication) at 60 days of age; 639 gilts completed the growth phase of the experiment that ended at 226 days of age.

Dams of project gilts were managed alike during the farrowing/lactation period. After weaning, all gilts were managed alike in the nursery until approximately 60 days of age (20.9 kg, 46 lb). They were then moved to the grow-finish facility where they were penned (10/pen) by line-treatment designation. They all were allowed ad libitum access to a corn-soybean meal-based diet and were managed alike until 123 days of age. A 3-phase growing-finishing diet was used: phase 1, 1.15% lysine (60 days to 36.3 kg, 80 lb); phase 2, 1.0% lysine (36.3 to 59 kg, 80 to 130 lb); and phase 3, 0.90% lysine (59 kg, 130 lb, to 123 days).

At 123 days, pens of gilts on the ad libitum regimen (AL) were allowed ad libitum access to a corn-soybean meal-based diet (0.70% lysine, 0.70% Ca, 0.60% P) until they were moved into the breeding barn. Gilts on the restricted intake regimen (R) received a corn-soybean meal-based diet at approximately 75% of the energy intake as AL-fed gilts until moved into the breeding barn. Energy restriction was achieved by predicting intake with a quadratic equation of average daily feed intake on body weight of AL-fed gilts. The predicted ad libitum intake (based on the projected body weight for the upcoming two-week period) was multiplied by 0.75 to determine the daily feed intake for R gilts. The diet contained 0.93% lysine, 1.0% Ca, and 0.80% P. All vitamins and minerals, except selenium, were increased so that daily intake of these nutrients per unit of body weight was expected to be equal for gilts on both diets. Additional details of the diets and management are in two articles in the *2007 Nebraska Swine Report* (Johnson et al, *2007 Nebraska Swine Report*; Miller et al., *2007 Nebraska Swine Report*).

Gilts were weighed and backfat and longissimus muscle area were recorded every 14 days until final measurements were recorded at an average age of 226 days. Beginning at approximately 140 days of age, gilts were moved by pen to an adjacent building where boar exposure and estrus detection occurred. Date of first observed estrus and each additional estrus were recorded.

Breeding and Lactation Management

Gilts in good health that could be mated at second or later postpubertal estrus during a predetermined breeding period were identified as breeders and moved to the breeding barn at approximately 230 days of age. Breeding commenced approximately 10 days later. A breeding period of 25 days (Rep 1), 24 days (Rep 2), 26 days (Rep 3), and 28 days (Rep 4) was used to match the unit's production schedule. Gilts were checked twice daily for estrus and inseminated each day that they were observed in estrus. Insemination was with semen from commercial terminal sire line boars. Gilts were in pens of approximately eight per pen until inseminated and then were moved to gestation stalls. Gilts that did not express estrus, those that were mated but diagnosed open with an ultrasound pregnancy test 50 days postbreeding, those that were diagnosed pregnant but did not farrow a litter, lame gilts, and gilts in poor health were culled.

While in the breeding barn and during gestation, all gilts were fed a standard corn-soybean meal-based diet (13.8% protein, 0.66% lysine) at the rate of 1.8 kg, 4.0 lb, daily until 90 days of gestation when feed intake was increased to 2.3 kg, 5.0 lb, daily. At approximately 110 days of gestation, females were weighed, scanned for 10^{th} rib backfat thickness, and placed in farrowing crates in rooms of 12 crates per room. They were fed 2.7 kg, 6 lb, per day of a corn-soybean meal-based lactation diet (18.5% protein, 1.0% lysine). Sows were provided only a small amount of feed on the day they farrowed, 2.7 kg, 6 lb, on the second day, 4.5 kg, 10 lb, the third day, and then were given ad libitum access to feed. The total number and number of live pigs in each litter were recorded. Pigs were fostered among litters without regard to line or gilt developmental regimen to reduce variation in number nursed per sow. Litters were weaned at an average age of 17 days and the number weaned and total litter weight were recorded. Weight and ultrasonic backfat of each sow at weaning was recorded. They were then moved to the breeding area and placed in pens of approximately eight sows per pen.

Feeding, estrus detection, insemination, and management during gestation and subsequent lactations were as described above for gilts. The breeding period for sows within replications and parities ranged from 24 to 32 days. Breeding continued until 10 days after the last sow in the replication was weaned. Every sow had at least 10 days to express postweaning estrus, and most had 15 to 20 days. Sows that did not express estrus, those that were detected to be open by ultrasonic pregnancy test, and those diagnosed pregnant but that did not farrow a litter were culled. Lame and unhealthy sows also were culled.

Traits and Data Analysis

Reproductive success is an all or none outcome; gilts and sows either did or did not produce litters. This outcome is a binomial trait that can

Table 1. Numbers of gilts from 60 to 230 days of age.

		outcome, day 0 to final test date						Culled, not breeders				
Line[a]	Trt[b]	N_{60}	$N_{Died, 60-123 days}$	$N_{123 days}$	$N_{died/FL/Rupt,123 to 226 days}$[c]	$N_{226 days}$	N_{AP}[d]	$N_{No AP}$	$N_{AP-late}$[e]	$N_{Died/FL/Rupt}$[f]	N_{Random}[g]	No Breeders
LW x LR	A	177	1	176	2	174	159	15	3	6	11	139
LW x LR	R	178	3	175	4	171	133	38	4	2	4	123
L45X	A	153	3	150	3	147	143	4	3	1	10	129
L45X	R	153	2	151	4	147	133	14	5	2	8	118
Total		661	9	652	13	639	568	71	15	11	33	509

[a]LW x LR = Large White x Landrace cross females, L45X = Line 45 cross females.
[b]A = ad libitum access to feed to breeding age (230 days), R = energy restriction (75% of A) from 123 to 230 days of age.
[c]Number that died or were removed from test for foot and leg problems, or were ruptured.
[d]Number that expressed pubertal estrus.
[e]Number that expressed pubertal estrus late in the development period, but were culled because they could not be mated at second postpubertal estrus.
[f]Number completing development period that died, ruptured, or were culled for foot/leg problems.
[g]Number that were culled randomly to reduce numbers to available breeding/farrowing spaces.

Table 2. Weight, backfat, and longissimus muscle area at 226 days of age, and age at puberty.[a,b]

	Weight, kg		Backfat, cm		Longissimus area, cm^2		Age at puberty, days	
Item	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
LW x LR	128.1	2.06	2.32	0.15	40.6	1.05	178.6	3.33
L45X	124.1	2.10	2.44	0.14	39.2	1.05	173.0	3.34
P-value	0.0025		0.048		0.002		0.003	
A	137.3	2.04	2.88	0.14	42.6	1.04	174.1	3.27
R	115.0	2.04	1.89	0.14	37.3	1.04	177.5	3.29
P-value	<.0001		<.0001		<.0001		0.017	

[a]LW x LR = Large White x Landrace cross females, L45X = Line 45 cross females.
[b]A = ad libitum access to feed to breeding age (230 days), R = energy restriction (75% of A) from 123 to 230 days of age.

be coded as 1 (success) or 0 (failure). Gilts completing the growth test were coded as 0 if they did not express a pubertal estrus and 1 if they did. Then, based on females designated for breeding, they were coded as 1 or 0 if they did or did not produce litters at each of parities 1 to 4. Each female received four scores. A gilt designated for breeding that did not produce a P1 litter received scores of 0, 0, 0, 0 for reproductive success at each parity. One that produced a P1 litter, but not a P2 litter, received scores of 1, 0, 0, 0; one that produced two litters received scores of 1, 1, 0, 0, etc., and one that produced four litters received scores of 1, 1, 1, 1. These scores measure reproductive success rates. They were fitted with general linear models designed for binomial data to determine the importance of line, gilt treatment, and interaction of line with treatment on reproductive rate through 4 parities. The effects of replication, sire, and litter of gilt were fitted in models as random effects to account for those sources of variation.

Number of live pigs per litter and number and total weight of pigs weaned by each sow were analyzed with models that included line, treatment, interaction, and random effects of replication, sire, and litter of gilt. Number of pigs that sows were given an opportunity to raise (number after crossfostering) and age at weaning were included as covariates to adjust all sows to a common number nursed and lactation length. Lifetime productivity of each female designated for breeding was calculated as the total number of live pigs, total number of weaned pigs, and total weight of pigs at weaning that she produced through parity 4. These measures of lifetime production were fitted to the same model as described above.

Results

Table 1 contains the numbers of pigs at each stage during the gilt developmental period. Of the total 661 gilts that started the experiment at 60 days of age, 9 died between 60 and 123 days and 13 died or were removed for structural or health reasons between 123 and 226 days of age. These losses were approximately equal across lines and treatments. Of the 639 gilts that completed the developmental period, 568 expressed a pubertal estrus by 230 days of age when gilts were moved to the breeding barn; 15 of these gilts were culled because they expressed estrus very late in this period and could not be mated at second or greater postpubertal estrus. Line and treatment affected both age at puberty and the proportion of gilts that expressed pubertal estrus (see below for results of analysis). Eleven gilts that expressed estrus either died or were culled for structural or health reasons before breeding, and an additional 33 gilts that qualified as breeders were culled at random to reduce the numbers to available breeding and farrowing spaces. A total of 509 gilts were designated as breeders, and it is these gilts for which lifetime production scores and productivity were recorded and analyzed.

The *2008 Nebraska Swine Report* contains articles summarizing effects of line and gilt developmental regimen on growth of gilts to 226 days of age and

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Table 3. Number of gilts that produced litters at each parity and numbers removed for reproductive failure, death, or unsoundness.[a]

		Parity 1							Parity 2						
Line	Trt	Brds	No. Lits	Mated open	Cull no est	Died gest	Cull FL/Inj	Died far	Brds	No. Lits	Mated open	Cull no est	Died gest	Cull FL/Inj	Died far
		Parity 1							Parity 2						
LW x LR	A	139	107	22	8	2	0	4	103	63	16	17	7	0	0
LW x LR	R	123	97	16	5	2	3	1	96	65	16	14	1	0	1
L45X	A	129	112	15	1	1	0	1	111	63	24	24	1	0	1
L45X	R	118	96	13	4	4	1	0	96	65	15	10	5	1	0
Total		509	412	66	18	9	4	6	407	256	71	65	14	1	2
		Parity 3							Parity 4						
LW x LR	A	63	53	8	2	0	0	0	54	41	12	1	0	0	0
LW x LR	R	64	51	10	2	1	0	0	51	41	8	1	1	0	0
L45X	A	62	51	6	4	0	1	0	51	44	7	0	0	0	0
L45X	R	65	56	5	3	1	0	1	55	44	7	3	1	0	0
Total		254	211	29	11	2	1	1	211	170	34	5	2	0	0

[a]Brds = Number of females designated as breeders; No. lits = number of litters; Mated open = number mated but culled because they were diagnosed as not pregnant; Cull no. est = number that were culled because they did not express estrus during the breeding period; Died gest = number died during gestation, Cull FL/Inj = number culled for foot and leg or other soundness condition; Died Far = number that died during farrowing.

Table 4. Probability of reproductive success.

	Pr Pub Estrus[a]		Pr P1 Litter[b]		Pr P2 Litter[b]		Pr P3 Litter[b]		Pr P4 Litter[b]	
Item	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
					Line means					
LW x LR	0.88	0.034	0.78	0.038	0.49	0.057	0.40	0.073	0.31	0.073
L45X	0.95	0.019	0.84	0.032	0.52	0.057	0.43	0.075	0.34	0.078
P-value	0.010		0.076		0.633		0.562		0.436	
					Treatment means					
A	0.96	0.016	0.83	0.034	0.47	0.056	0.38	0.072	0.31	0.073
R	0.86	0.037	0.80	0.037	0.54	0.057	0.44	0.075	0.34	0.078
P-value	0.0001		0.521		0.135		0.203		0.386	

[a]Probability that gilts that completed the development period expressed pubertal estrus by 225 days of age.
[b]Probability that gilts identified as breeders produced parity 1, 2, 3, and 4 litters.

age at puberty. To illustrate effects of line and treatment on growth, mean final weight, backfat, longissimus muscle area, and age at puberty are in Table 2. Lines responded similarly to treatment as interaction of line by treatment was not significant for any trait. LW x LR gilts were 4 kg heavier with 0.12 cm less backfat and 1.4 cm^2 greater LMA than L45X gilts (all $P < 0.05$). L45 X gilts were 5.6 days younger ($P < 0.01$) at puberty than LW x LR gilts. Treatment effects were significant for all traits. Developing gilts with 25% energy restriction caused them to weigh 22.4 kg less, to have .99 cm less backfat and 5.3 cm^2 less LMA, and to be 3.4 days older at puberty than developing them with ad libitum access to feed.

Table 3 contains the number of females that produced litters at each parity, number of sow deaths, and the number of sows culled for reproductive failure, health and structural conditions. Most sow losses were those that were mated and subsequently returned to estrus. Overall, the percentages of breeding females that fit that category were 13, 17, 11, and 16% at parities 1 to 4, respectively. The incidence of culling for return to estrus after insemination during the breeding period did not differ significantly between lines or treatments, although it was somewhat greater for LW x LR than L45X sows (16 vs. 13%) and for gilts developed with restricted energy intake than those fed ad libitum (13.5 vs. 10.3%).

The second most frequent cause of culling sows was failure to express estrus during the breeding period, which includes those that may have expressed an estrus that was not detected by technicians. The incidence of that condition was low in gilts (3.5%), quite high for parity 1 sows being mated for parity 2 litters (16%), and low again for mating of parity 2 and 3 sows for their next litter (4.3 and 2.4%, respectively).

In many production herds, sows that return to estrus after insemination are often inseminated again and given another chance to reproduce, and some with delayed returns to estrus are eventually inseminated and conceive. These practices would lead to more lit-

Table 5. **Mean number of live pigs, number weaned, and litter weaning weight for females with litters, per female with litter, and lifetime productivity per gilt designated for breeding.**

			Number of live pigs		Number weaned		Litter weaning weight, kg	
Line[a]	Trt[b]	Parity	Mean	SEM	Mean	SEM	Mean	SEM
LW x LR			11.73	0.31	10.01	0.11	55.3	0.9
L45X			12.13	0.31	9.76	0.11	52.8	0.9
	A		11.90	0.31	9.82	0.11	53.4	0.9
	R		11.96	0.31	9.95	0.11	54.7	0.9
		1	11.74	0.30	10.08	0.10	50.5	0.9
		2	11.53	0.32	10.20	0.12	57.8	1.0
		3	12.20	0.33	9.95	0.13	55.9	1.0
		4	12.24	0.35	9.32	0.14	51.9	1.1
P-values for line, treatment, and parity effects								
Line			0.12		0.05		0.008	
Trt			0.81		0.27		0.13	
Parity			0.02		<.0001		<.0001	
Lifetime production per gilt designated for breeding								
LW x LR			23.37	3.32	19.48	2.69	106.2	16.3
L45X			26.05	3.33	21.12	2.70	112.9	16.3
	A		23.83	3.30	19.60	2.69	105.4	16.2
	R		25.59	3.32	21.00	2.71	113.8	16.3
P-values for lifetime production								
Line			0.14		0.25		0.41	
Trt			0.31		0.32		0.28	

[a]LW x LR = Large White x Landrace cross females; L45X = Line 45 cross females.
[b]A = ad libitum access to feed to breeding age (230 days); R = energy restriction (75% of A) from 123 to 230 days of age.

ters from the same set of females than in this experiment, in which these sows were all culled and not given another chance to reproduce. To ensure a uniform culling policy for all replications, parities, lines, and treatments, we determined that those sows would be culled. Also, giving the sows additional opportunities to reproduce extends the farrowing period, which did not fit our production schedule.

Of the 509 gilts designated for breeding, 27 (5.3%) died during one of the four gestation periods and 9 (1.8%) died during farrowing. Very few sows were culled for foot and leg or health conditions.

Table 4 contains results of statistical analysis of the 0/1 binomial scores for reproductive success. The analysis produced the probability statistics in the table. The first of these is the probability that a gilt that finished the development period expressed estrus by 230 days of age. Both line and treatment significantly affected this probability; 95% of L45X gilts reached puberty compared with 88% of LW x LR gilts ($P = 0.01$), and 96% of gilts developed with ad libitum intake reached puberty compared with 86% of gilts developed with energy restriction ($P = 0.0001$). Gilts of both lines responded similarly to treatments as there was no interaction.

Probabilities of females producing parity 1 to 4 litters are all based on gilts designated as breeders at 230 days of age. No effect, line, treatment, or interaction, was significant for any of these probabilities. The greatest difference was between lines for the probability that gilts designated as breeders produced a parity 1 litter, being .84 for L45X gilts and .78 for LW x LR gilts ($P = 0.076$). The probability that females produced parity 2, 3, and 4 litters was greater for those developed with restricted energy intake, but none of these differences approached statistical significance ($P > .10$).

Although not significant, line differences in this experiment are consistent with differences observed in previous comparisons. Line 45X females had 0.4 more live pigs per litter than LW x LR females, but their maternal abilities were not as good. When given an opportunity to raise the same number of pigs, LW x LR females weaned 0.25 more pigs per litter and total litter weight was 2.5 kg more than for L45X females. Gilt development regimen had almost no effect on subsequent litter size or maternal ability in either line as interaction of line and treatment was not significant.

All measures of lifetime production were greater for L45X females than LW x LR females and for females developed with restricted energy intake (Table 5). However, none of these measures, total number of live pigs at birth, total number weaned, nor total weight of litter weaned, all calculated per gilt designated for breeding, was significantly affected by line, treatment, or interaction. Lifetime sow productivity is a difficult trait to evaluate experimentally. The observed differences were relatively large, ranging from 8 to 11%, and if real, are economically important. Yet, in an experiment in which 509 females produced 1,049 litters and in which variation was controlled and culling criteria strictly adhered to, natural variation was still large enough that observed differences could be explained by chance as all *P*-values were > = 0.14.

Thus, we conclude from this project that prolific gilts that differ in rate of lean growth respond similarly to a developmental regimen in which energy intake from 123 days of age to breeding was restricted to 75% of that of gilts developed with ad libitum intake. Further, this energy restriction decreased the proportion of gilts that had expressed estrus by 230 days of age and increased the age at puberty for those that did express pubertal estrus. Thereafter, females developed with both regimens had similar reproductive performance at each parity and similar lifetime production.

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An Economic Analysis of Energy Restriction During Pre-Pubescence in Gilts

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Summary

This report evaluates the economic costs of production and profits for energy-restricted and conventional gilt development programs. Production and performance data and input and output prices were used to construct enterprise budgets for both groups. Results indicate that restricting feed intake during gilt development lowered breakeven selling prices for market pigs by an average of $0.17/cwt for two prolific maternal lines through their first four parities.

Introduction

The traditional method of developing breeding gilts is to provide feed on an ad libitum basis until they are bred. The conventional reasoning behind self-regulation of feed intake is to allow gilts to grow as fast as possible to hasten the onset of puberty because more mature animals typically have a greater likelihood of successful conception; thus, this method has been viewed as both production and profit enhancing. However, body weight has not been conclusively shown to affect age at puberty. Kirchgessner et al. (1984) reported that reducing energy intake to 70 to 75% of ad libitum intake did not affect age of first estrus, while Le Cozler et al. (1998) reported that gilts fed to 80% of full intake had a later first-detected estrus than gilts fed to appetite. However, in the Le Cozler study, age of service was not different between control and energy-restricted gilts. Additionally, this conventional process may result in an increased probability of overweight gilts and, therefore, the possibility for lower production due to breeding difficulties. Increased body weights could also cause mobility problems later in life, leading to increased culling rates or even death losses, both of which can negatively affect profits.

In a multiyear study, Johnson et al. (*2005-2008 Nebraska Swine Reports*) focused on reducing these production inefficiencies by restricting energy intake to 75% of ad libitum for two prolific maternal lines of gilts from 123 to approximately 226 days of age. This restricted energy development program should result in less feed and feed expense compared to conventionally developed gilts, but it also may lead to more late maturing gilts that have to be culled, thus increasing overall development costs. Therefore, economic differences between conventional and restricted energy development programs are not clear. This project develops an enterprise budget for each development program to determine the relative profitability of each system over four parities.

Materials and Methods

An enterprise budget was created to estimate revenue and costs using production data from the Johnson et al. studies. The unit of measurement for the budget was an individual sow and the budget was organized into three main sections: gilt development, nursery and market pig production for the first four parities, and an output page summarizing the revenues and costs for the sow and her market pigs throughout their lifetime. Included in the development section are production parameters (e.g., average daily gain, feed intake, initial weight, ending weight, days spent in the development program, etc.), cull credits, building and equipment costs, interest, veterinary expense, utilities, labor, and ration composition. The nursery and market pig production section includes production parameters similar to those in the development section (e.g., average daily gain, feed intake, etc.), but also includes 3 sow and 12 market pig feed rations, sow and market-pig cull credits, fixed costs for both the breeding sow and her offspring, interest cost, income, fixed costs, and additional variable costs (Table 1). Parity-specific results are then reported in the form of breakeven selling price for progeny and in total profit/loss. In the summary section of the budget, all costs and credits are summed and results are reported both as total profit/loss earned per sow for each treatment and in the form of breakeven selling price of market pigs by treatment.

The maternal lines used in the study were Large White-Landrace (LWxLR) and Nebraska Line 45 cross (L45X) described in the previous report (Johnson et al. 2010). The LWxLR and L45X gilts were half-sibling as they were produced by dams that were artificially inseminated with semen of the same industry maternal line boars. Production records including number of pigs weaned, weaning weights, lactation feed intake, etc. through four parities were kept for each sow. These production data, along with input and output prices and other production assumptions, were used to construct the budgets. Feed costs were calculated using typical ration compositions based on NRC requirements and 2004-06 historical average prices were used for input and output prices (Table 1). Fixed costs,

Table 1. Input and output prices per unit.[1]

Item	Price per Unit	Unit	Type of Expense	Cost per Pig	Cost per Litter
Market Swine Selling Price[2]	$0.43	/lb	Veterinary and Health Cost	$4.72	
Corn	$2.14	/bu	Utilities	$1.57	
Soybean Meal	$200.08	/ton	Marketing and Transportation Costs	$1.68	
Tallow	$0.29	/lb	Other Misc. Costs		$10.00
Dicalcium Phosphate	$0.22	/lb	Labor		$62.28
Limestone	$0.02	/lb	Annual Fixed Costs (per pig-space)	$18.24	
Salt	$0.07	/lb	Annual Fixed Costs (per sow-space)		$79.30
			Breeding Costs		$20.00

[1]Prices from 2004-06.
[2]Liveweight basis.

Table 2. Event probabilities for two prolific maternal lines.

	Line			
	LWxLR		L45X	
Outcome	Ad Libitum	Restricted	Ad Libitum	Restricted
Parity 1 Litter	0.7714	0.7910	0.8695	0.8152
Parity 2 Litter	0.4581	0.5298	0.4846	0.5477
Parity 3 Litter	0.3848	0.4140	0.3841	0.4697
Parity 4 Litter	0.2888	0.3265	0.3242	0.3610

Table 3. Revenue and cost of production for two prolific maternal lines.

	Line					
	LWxLR			L45X		
Item	Ad Libitum	Restricted	Difference[1]	Ad Libitum	Restricted	Difference[1]
Total cwts Produced (per sow)[2]	46.41	51.35	4.94	49.84	52.81	2.97
Revenue (per sow)	$2,008.75	$2,222.51	$213.76	$2,157.25	$2,286.02	$128.77
Gilt Production (per gilt)						
Variable Costs	$123.59	$115.11	($8.48)	$120.29	$113.08	($7.21)
Fixed Costs	$6.91	$7.67	$0.76	$6.46	$7.02	$0.56
Total Costs	$130.50	$122.78	($7.72)	$126.75	$120.09	($6.66)
Market Swine (per litter through 4 parities)						
Variable Costs	$1,411.31	$1,566.71	$155.40	$1,526.61	$1,628.85	$102.24
Fixed Costs	$253.64	$280.63	$26.99	$272.39	$288.65	$16.26
Total Costs	$1,664.95	$1,847.34	$182.39	$1,799.00	$1,917.50	$118.50
Total Cost (per sow)	$1,795.45	$1,970.12	$174.67	$1,925.75	$2,037.59	$111.84
Profit/Loss (per sow)	$213.30	$252.39	$39.10	$231.49	$248.43	$16.93
Breakeven Selling Price (per cwt)[2]	$38.69	$38.37	($0.32)	$38.64	$38.58	($0.06)

[1]Restricted minus *Ad Libitum*.
[2]Liveweight basis.

veterinary expense, transportation costs, utilities, breeding cost and amount of labor are from Lawrence and Ellis (Iowa Estimated Monthly Returns from Farrowing and Finishing Hogs; Table 1). An agricultural labor wage rate of $10.53/hour is from the National Agricultural Statistics Service.

Profits and breakeven selling prices were calculated by finding the revenue, fixed cost, variable cost, and total cost for each possible scenario which varied according to length of time the gilt/sow remained in the program before being culled. These outcomes were gilt development, gilt through first parity of market pigs, gilt through second parity, gilt through third parity, and gilt through fourth parity. The probability of each of these outcomes was used to determine the weighted average revenue, costs, and profit for an average gilt entering the program. Because gilts from each treatment and line had different probabilities of successfully farrowing each of the four parities, different probabilities were used for each treatment, line, and parity. These probabilities are summarized in Table 2. For example, ad libitum LWxLR gilts have a cost of gilt development of $149.63 ($153.78 from Table 3 plus breeding costs and subtracting cull credits) and a cost of first, second, third, and fourth parity litters of approximately $883.85, $924.93, $826.74, and $724.47 (fourth parity cost includes a credit for selling value of sow), respectively. The probabilities of these outcomes occurring are 1, 0.7714, 0.4581, 0.3848, and 0.2888, respectively. Multiplying the probability of each outcome by each of the cost components in the budget and summing those products results in a total cost of $1,796.64.

Results

Results for each line and treatment are summarized in Table 3

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in the form of revenue, fixed costs, variable costs, and total costs for gilt development and market pig production. Energy-restricted gilts were more productive than nonrestricted females as they produced an average of 5.12 more cwts per developed LWxLR gilt (48.05 cwts sold per ad libitum gilt vs. 53.17 cwts sold per restricted gilt; Table 3) and 2.97 more cwts per developed L45X gilt (49.84 cwts sold per ad libitum gilt vs. 52.81 cwts sold per restricted gilt; Table 3). The increased production was primarily caused by energy-restricted females having a greater probability of farrowing a litter than an ad libitum gilt at each parity. An average energy-restricted LWxLR gilt had a greater probability of farrowing first, second, third, and fourth parity litters than ad libitum females. Contrary to LWxLR gilts, an average energy-restricted L45X gilt did not have a greater probability of farrowing a first parity litter, but did have a greater probability of farrowing second, third, and fourth parity litters than an average ad libitum gilt. However, in no case were these differences statistically significant. Additionally, as selling price increases, energy restriction during gilt development becomes more economically advantageous because, as previously mentioned, energy-restricted gilts produced a greater number of hundredweights than ad libitum gilts.

In addition to being more productive, limit-fed gilts were also less expensive to produce by an average of $9.74 for LWxLR females ($153.78 per ad libitum gilt vs. $144.04 per restricted gilt; Table 3) and $7.58 per L45X gilt ($149.59 ad libitum vs. $142.01 restricted; Table 3). Although fixed costs were $0.73 greater per gilt for restricted LWxLR females ($6.64 ad libitum vs. $7.37 restricted; Table 3) and $0.53 per gilt more expensive for restricted L45X gilts ($6.21 ad libitum vs. $6.74 restricted; Table 3), this was more than offset by the large reduction in variable costs for energy restricted females (Table 3). Variable costs are lower because energy-restricted females consumed less feed than their ad libitum counterparts.

On average, progeny from restricted fed LWxLR gilts had a $0.47/cwt lower breakeven selling price than ad libitum market pigs ($37.39/cwt ad libitum vs. $36.92/cwt restricted; Table 3). However, progeny from energy-restricted L45X dams had a $0.14/cwt higher breakeven selling price than progeny from nonrestricted dams ($37.78/cwt ad libitum vs. $37.92/cwt restricted; Table 3). The lower breakeven selling price can be attributed to the increased production of energy-restricted gilts and also to the lower feed cost of limit feeding gilts during development.

The results from the budget analysis make sense intuitively. For instance, market swine production costs were greater for energy-restricted gilts from both genetic lines because they produced a larger number of offspring. One peculiar result, which was seemingly contradictory, was the greater profit and higher breakeven selling price in the L45X genetic line. One would assume the group with the lower breakeven selling price of progeny would also correspond to a greater profit or a lower loss. However, because restricted gilts produced a greater number of progeny, the magnitude of the profit/loss generated by the restricted gilt is greater than that of the ad libitum. To reiterate what was said previously, when profits are large for an average ad libitum gilt, they are greater for an average restricted gilt and when losses are large for an ab libitum gilt, they also are larger for a restricted gilt because of the increased reproductive production.

One important caveat to this research was an increased rate of culled animals during the development stage when restricting energy in developing gilts. Because of this, a greater number of gilts at the beginning of the program would be needed, leading to larger fixed costs incurred per developed gilt. As previously mentioned, this increase in fixed costs is more than offset by the decrease in feed costs when restricting energy, but could have practical implications for swine producers as more barn space would be needed to produce the same number of breeding gilts as the traditional method of gilt development.

These results have important implications for swine producers as restricting energy intake for breeding gilt production did not adversely affect sow productivity. The savings of feed costs counteracted the negative aspects of energy restriction in gilt development (increased rate of culling during development, etc.). Additionally, producing breeding gilts approximately $8.66/head cheaper, which was the average difference in energy-restricted females, reduced progeny breakeven selling prices in this study by an average of approximately $0.17/cwt sold. Although this cost-savings is small, swine production is a low-margin industry where saving pennies per cwt are essential to a successful business.

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Effects of Distillers Grains With Solubles (DDGS) and Paylean® Supplementation on Carcass Quality, Color Stability, and Sensory Characteristics of Pork

Withdrawing dietary DDGS four weeks prior to harvesting partially alleviated the reduced saturated and increased unsaturated fatty acid concentrations in fat observed due to DDGS feeding during growing and early finishing periods. Dietary inclusion or withdrawal of DDGS and RAC does not affect chemical composition and minimally affected sensory characteristics of pork.

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Summary

Forty pigs (66.6 lb) were used in a 14-week 4-phase regime study conducted to evaluate the effect of feeding varying concentrations of DDGS to growing-finishing pigs formulated on a standardized ileal digestibility (SID) lysine (lys) basis, DDGS withdrawal at the last feeding phase, and ractopamine (RAC) supplementation 4 weeks prior to harvesting on carcass quality, and color stability and sensory characteristics of longissimus muscle (LM) of finishing pigs. Treatments consisted in 0, 15, or 40% dietary DDGS inclusion supplemented with or without RAC (4.5 ppm) 4 weeks prior harvesting. Final body weight, hot carcass weight, and dressing percentage were not affected by dietary DDGS inclusion, withdrawal or RAC supplementation ($P > 0.10$). Color characteristics were not affected by dietary DDGS inclusion or withdrawal ($P > 0.10$); however, dietary RAC supplementation reduced a and b* at days 0 and 7 ($P > 0.10$). Total polyunsaturated fatty (TPUFA) acids increased and total saturated fatty acids (TSFA) were decreased in response to increased dietary DDGS inclusion ($P < 0.01$); however, DDGS withdrawal partially alleviated these changes in fatty acid composition by increasing TSFA and reducing TPUFA ($P < 0.01$). The inclusion of RAC decreased TSFA and increased total monounsaturated fatty acids concentration ($P = 0.03$ and 0.04, respectively). Sensory characteristics were not affected by dietary RAC, DDGS inclusion or DDGS withdrawal ($P > 0.10$). The results of this investigation suggest that dietary RAC, DDGS inclusion or DDGS withdrawal did not affect carcass quality as evaluated by color, chemical composition, and sensory characteristics of LM of growing-finishing pigs. Increasing the concentration of dietary DDGS altered the fatty acid profile of backfat of pigs by decreasing saturated and increasing unsaturated fatty acids. However, withdrawing DDGS, 4 weeks prior to harvesting partially alleviated the increase in PUFA, and consequently the "soft pork" problems associated with the use of DDGS.*

Introduction

Evidence available in the literature indicates that dietary DDGS inclusion greater than 30% can be used in diets for growing-finishing without negatively affecting growth performance; however, the effect of dietary inclusion of DDGS may result in altered carcass characteristics and pork quality. Among the most important effects of dietary inclusion of DDGS on swine diets is the altered fatty acid profile of adipose tissue. Evidence indicates that inclusion of ractopamine (RAC) may affect carcass characteristics and pork quality especially by increasing protein and reduced fat deposition. Research has been conducted to reduce the changes in carcass characteristics originated by the dietary DDGS inclusion. Among other strategies, DDGS withdrawal during the late-finishing phase has been used to alleviate the negative effect of dietary DDGS inclusion on carcass characteristics. Ractopamine addition also may help to alleviate problems associated with the unsaturated fat content of DDGS by reducing fatty acid deposition. This report is a companion article to a previous article in the *2009 Nebraska Swine Report* in which the feeding value of diets for growing-finishing pigs with varying DDGS concentration, DDGS withdrawal, and RAC inclusion was reported. The present report examines the effect of dietary DDGS concentrations of 15 and 40% and the interaction with the inclusion of RAC, DDGS withdrawal, or both during the last 4 weeks of the finishing period on carcass characteristics, color stability, and sensory characteristics of pork.

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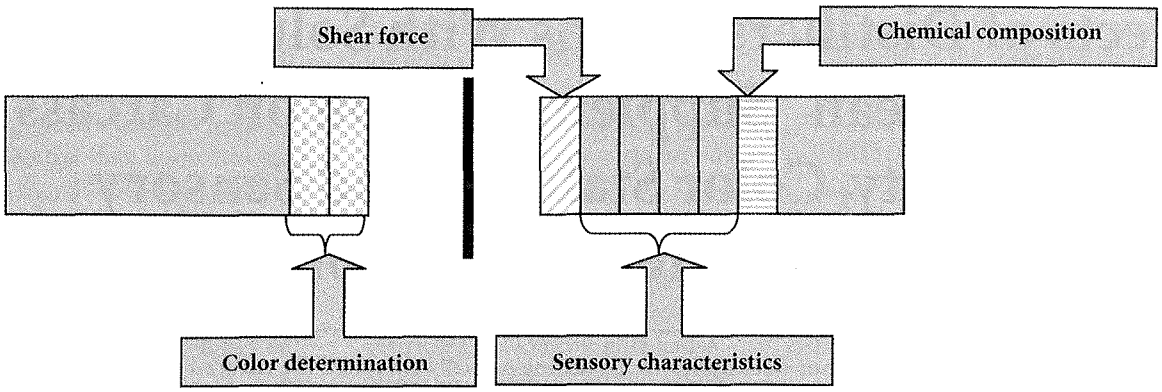

Figure 1. Longissimus muscle sections of the loins used for shear force, color determination, sensory characteristics, and chemical analysis.

Materials and Methods

Carcass Data Collection

Forty barrows (weighing an average of 66.6 lb at the beginning and 273.2 lb at the end of the feeding period) were assigned to 4 dietary regimens designed to provide DDGS inclusion of 0, 15 or 40% throughout the experiment or 40% dietary DDGS inclusion during the first 3 feeding phases and 0% dietary DDGS inclusion during the last feeding phase. Eight treatments were produced by randomly assigning pigs to 1 of 4 dietary treatments or their RAC-supplemented counterparts. Details of the growth study are described in a companion article (*2009 Nebraska Swine Report*). At the end of the feeding phase, all pigs were transported to a commercial pork packing facility located approximately 170 miles from the University of Nebraska Swine Research Unit. Pigs were weighed before entering (live weigh; LW) and before leaving the harvesting floor (hot carcass weight; HCW). Dressing percentage (DP) was calculated using the following formula DP = ((LW / HCW) × 100).

Carcasses were subjected to a standard spray-chilling procedure for 24 hours. Before entering the fabrication floor, backfat samples were obtained (perpendicular to the 10th rib), submerged in liquid nitrogen and maintained at -112°F until analyzed for fatty acid profile. Carcasses were identified on the chilling floor, marked in the vertebrae, and the bone-in loin (410 pork loin; NAMP, 1997. The Meat Buyer Guide. North American Meat Processors Association. Reston, Va.) from the right side of the carcass was collected. The collected loins were individually vacuum packed and transported to the Meat Science Laboratory at the University of Nebraska for further analysis. Seven days postmortem the loins were boned and a section of longissimus muscle (LM; 412B pork loin, boneless, center-cut, eight ribs; NAMP, 1997. The Meat Buyer Guide. North American Meat Processors Association. Reston, Va.). Nine 1-inch sections (Figure 1) were obtained and used for color determination, shear force estimation, sensory characteristics evaluation, and chemical composition.

Color Determination

The two sections of the LM used for color determination were packed in Styrofoam trays, wrapped with PVC film, and maintained at 34°F under fluorescent light illumination for 7 days. Color spectrometry measurements L*, a*, and b* (representing lightness, redness, and yellowness, respectively) were obtained through the packing film on five sites on each section at the beginning (day 0) of the 7-day color experiment and daily thereafter using a Hunter Lab® Mini Scan XE plus (Model 45/0-L, Reston, Va.) handheld colorimeter. The calibration of the colorimeter was performed using black and white tiles. The change in total color (E) was calculated as $[((L^* \text{ at d } 10 - L^* \text{ at d } 0)^2 + (a^* \text{ at d } 10 - a^* \text{ at d } 0)^2 + (b^* \text{ at d } 10 - b^* \text{ at d } 0)^2)^{1/2}$; Minolta, 1998. Precise color communication-color control from perception from instrumentation. Minolta Corp., Ramsey, N.J.]. This formula was developed in order to better describe the changes in color that would occur during periods of retail display.

Warner-Bratzler Shear Force Analysis

The loin sections used for Warner-Bratzel shear force (American Meat Science Association. Research guidelines for cookery, sensory evaluation, and tenderness measurements of meat. 1995) were vacuum-packed and maintained at -4°F until analysis. Before the analysis, chops were allowed to thaw, cooked to an internal temperature of 158°F on a Hamilton Beach® grill (Washington, N.C.), and cooled for four hours at 35.6°F. During the cooking process, temperature was monitored using thermocouples. Three cores of 0.5 in^2 from each section were removed parallel to the arrangement of the muscle fiber. Cores were sheared parallel to the muscle fiber using an Intron Universal Testing Machine (Model 55R1123, Canton, Mass.) equipped with a Warner-Bratzler shear attachment. The speed for the test was 250 mm/min.

Fatty Acid Profile

Fatty acid concentration was measured in the backfat of all pigs. Fatty acids were extracted in hexane and methyl esters were formed. The

Table 1. Attribute, magnitude and description and scale of sensory characteristics.

	Magnitude		
Attribute	0 mm	150 mm	Comments
General appearance	Very nonuniform	Very uniform	Color of interior meat
Toughness	Very tough	Very tender	During the first bite
Chewiness	Very hard to breakdown	Very easy to breakdown	During chewing
Juiciness	Very dry	Very moist	
Pork flavor	Lacking	Intense	
Off-flavor	Lacking	Intense	
Aftertaste pork flavor	Lacking	Intense	
Overall acceptability	Very undesirable	Very desirable	

mass ratio of fatty acids were quantified using a gas chromatograph (Hewlett-Packard, Model 5890, Farmington Hills, Mich.).

Sensory Evaluation

Loin sections used for sensory evaluation were vacuum packed and maintained at -4°F until further analysis. Chops were thawed, cooked, and sensory evaluation was conducted using 38 consumer panelists recruited from the Animal Science Department and the Department of Food Science and Technology at the University of Nebraska–Lincoln. The chops were cooked using an electric grill to an internal temperature of 158°F, and excess fat was trimmed. Samples of 1 in^2 were obtained and maintained warm until served to the panelists. Panelists used a descriptive unstructured line-scale to evaluate the attributes provided in Table 1.

Statistical Analysis

Carcass characteristics, chemical composition, fatty acid profile and sensory characteristics were analyzed as a complete randomized design using the MIXED procedure (SAS Inst., Inc., Cary, N.C.). Each pig was considered an experimental unit and pen was considered a random effect. Color data were analyzed as repeated measures in time using the MIXED procedure. Contrasts were designed to evaluate linear and quadratic responses to dietary DDGS inclusion and withdrawal as well as RAC inclusion. For the color stability study, pig was considered the experimental unit and tray was considered a random effect.

Results and Discussion

Carcass traits are shown in Table 2. Treatment did not affect hot carcass weight ($P = 0.54$), similarly, no effects of RAC or DDGS withdrawal were detected ($P = 0.56$ and 0.29, respectively). In contrast to results reported in the literature, DP was not affected by RAC inclusion ($P = 0.56$). In the present study, DP did not show a linear reduction in response to increasing

(Continued on page 15)

Table 2. Response and effect of dietary distillers grains with soluble (DDGS) inclusion and ractopamine (RAC) on carcass characteristics and chemical composition of longissimus muscle of growing-finishing pigs.

Treatment	1	5	2	6	3	7	4	8						
DDGS, % for G1, G2, and F1[a]	0	0	15	15	40	40	40	40						
DDGS, % for F2[b]	0	0	15	15	40	40	0	0						
RAC, ppm	0	4.5	0	4.5	0	4.5	0	4.5						
									P-value					
Item									SEM[c]	Trt[d]	L[e]	Q[f]	RAC	W[g]
No. of pigs	5	5	5	5	5	5	5	5						
Final BW, lb	272.52	272.9	289.7	306.4	271.2	271.8	279.5	287.3	12.08	0.50	0.31	0.70	0.46	0.33
Chemical composition, %														
Crude protein	21.61	22.49	21.97	22.35	21.38	21.63	21.37	21.98	0.50	0.57	0.56	0.71	0.12	0.71
Moisture	67.09	67.52	67.00	68.10	66.40	65.86	67.05	67.32	1.10	0.87	0.87	0.67	0.66	0.31
Ash	1.16	1.11	1.17	1.20	1.14	1.10	1.10	1.14	0.46	0.72	0.40	0.33	0.77	0.99
Fat	9.37	8.57	9.44	8.23	10.87	10.00	11.68	9.65	1.93	0.67	0.63	0.42	0.28	0.88
Carcass characteristics														
Hot carcass wt, lb	202.2	200.8	216.5	225.8	197.9	201.2	207.3	212.2	10.73	0.54	0.48	0.89	0.56	0.29
Dressing, %	74.15	73.51	74.66	73.74	72.88	73.94	74.08	73.81	0.53	0.48	0.12	0.21	0.61	0.32
10th rib BF, cm	3.60	3.60	4.20	3.75	3.40	3.80	3.60	3.60	0.52	0.96	0.87	0.53	0.97	1.00

[a]G1 = Grower1; G2 = Grower 2; F1 = Finisher 1.
[b]F2 = Finisher 2.
[c]SEM = Standard error of the mean.
[d]TRT = Treatment.
[e]L = Linear.
[f]Q = Quadratic.
[g]W = Withdrawal ; W × RAC interaction, $P > 0.05$.

Table 3. Response and effect of dietary distillers grains with solubles (DDGS) inclusion and ractopamine (RAC) on color, and shear force longissumus muscle of growing-finishing pigs.

Treatment	1	5	2	6	3	7	4	8
DDGS, % for G1, G2, and F1[a]	0	0	15	15	40	40	40	40
DDGS, % for F2[b]	0	0	15	15	40	40	0	0
RAC, ppm	0	4.5	0	4.5	0	4.5	0	4.5

									P-value					
Item									SEM[c]	Trt[d]	L[e]	Q[f]	RAC	W[g]
No. of pigs	5	5	5	5	5	5	5	5						
Shear force, lb	9.70	10.18	9.26	9.26	9.61	11.02	7.49	9.56	1.19	0.54	0.52	0.39	0.20	0.10
Color (day 0)														
a* (redness)	10.08	9.85	9.96	8.17	10.28	8.52	9.58	8.79	0.67	0.16	0.29	0.29	0.01	0.71
b* (yellowness)	15.40	14.46	15.44	13.89	15.05	12.82	14.27	13.64	0.63	0.04	0.62	0.49	0.01	0.97
L* (lightness)	52.12	48.10	51.8	49.54	50.15	47.57	50.05	51.35	1.25	0.13	0.38	0.78	0.04	0.15
Color (day 7)														
a* (redness)	12.23	13.26	11.39	13.19	12.77	12.33	13.02	13.12	0.61	0.27	0.46	0.25	0.12	0.35
b* (yellowness)	15.20	14.92	14.69	14.37	14.90	13.18	14.42	14.90	0.64	0.34	0.75	0.79	0.02	0.29
L* (lightness)	54.77	50.56	54.72	50.96	53.40	49.90	53.39	53.24	1.39	0.05	0.17	0.30	0.01	0.19
E[h]	3.54	4.73	3.46	5.64	4.28	4.96	4.84	5.00	0.64	0.16	0.16	0.28	0.01	0.61

[a]G1 = Grower1; G2 = Grower 2; F1 = Finisher 1.
[b]F2 = Finisher 2.
[c]SEM = Standard error of the mean.
[d]TRT = Treatment.
[e]L = Linear.
[f]Q = Quadratic.
[g]W = Withdrawal; W × RAC interaction, $P > 0.10$.
[h]Change of color.

Table 4. Response and effect of dietary distillers dried grains with solubles (DDGS) inclusion and ractopamine (RAC) on fatty acid profile of backfat of growing-finishing pigs.

Treatment	1	5	2	6	3	7	4	8
DDGS, % for G1, G2, and F1[a]	0	0	15	15	40	40	40	40
DDGS, % for F2[b]	0	0	15	15	40	40	0	0
RAC, ppm	0	4.5	0	4.5	0	4.5	0	4.5

									P-value					
Item									SEM[c]	Trt[d]	L[e]	Q[f]	RAC	W[g]
No. of pigs	5	5	5	5	5	5	5	5						
Fatty acid, mass %														
Myristic, (14:0)	1.31	1.24	1.23	1.20	1.18	1.07	1.21	1.23	0.05	0.15	0.14	0.81	0.20	0.05
Palmitic, (16:0)	24.60	23.05	22.34	22.22	21.54	19.64	22.74	22.52	0.68	<.01	<.01	0.55	0.02	<.01
Palmitoleic, (16:1)	2.12	1.91	1.82	1.85	1.63	1.62	1.78	1.93	0.11	0.01	0.02	0.34	0.88	0.01
Stearic, (18:0)	13.97	13.21	12.36	12.03	11.39	9.92	13.14	12.15	0.81	0.01	0.01	0.72	0.07	<.01
Oleic, (18:1)	40.93	42.67	39.77	42.54	38.21	38.87	39.97	39.53	0.98	<.01	0.83	0.13	0.05	0.13
Linoleic, (18:2)	9.28	10.64	14.54	13.46	18.79	21.04	14.52	14.34	0.83	<.01	<.01	0.08	0.24	<.01
α-linolenic, (18:3)	0.36	0.45	0.50	0.47	0.52	0.61	0.46	0.50	0.03	<.01	<.01	0.19	0.02	<.01
Others	7.39	6.79	7.40	6.18	6.70	7.19	6.14	7.75	0.76	0.50	0.52	0.76	0.87	0.99
TSFA[h]	39.89	37.54	35.94	35.46	34.12	30.65	37.10	35.91	1.43	<.01	<.01	0.92	0.03	<.01
TMUFA[i]	43.05	44.59	41.59	44.40	39.84	40.49	41.76	41.47	0.96	<.01	0.60	0.10	0.04	0.07
TPUFA[j]	9.65	11.09	15.04	13.94	19.32	21.65	14.99	14.84	0.86	<.01	<.01	0.08	0.22	<.01

[a]G1 = Grower1; G2 = Grower 2; F1 = Finisher 1.
[b]F2 = Finisher 2.
[c]SEM = Standard error of the mean.
[d]TRT = Treatment.
[e]L = Linear.
[f]Q = Quadratic.
[g]W = Withdrawal; W × RAC interaction, $P > 0.10$.
[h]Total saturated fatty acids.
[i]Total mono-unsaturated fatty acids.
[j]Total poly-unsaturated fatty acids.

Table 5. **Response and effect of dietary distillers dried grains with solubles (DDGS) inclusion and ractopamine (RAC) on sensory characteristics of longissumus muscle of growing-finishing pigs.**

Treatment	1	5	2	6	3	7	4	8						
DDGS, % for G1, G2, and F1[a]	0	0	15	15	40	40	40	40						
DDGS, % for F2[b]	0	0	15	15	40	40	0	0						
RAC, ppm	0	4.5	0	4.5	0	4.5	0	4.5						
									P-value					
Item									SEM[c]	Trt[d]	L[e]	Q[f]	RAC	W[g]
No. of pigs	5	5	5	5	5	5	5	5						
Attribute[h], mm														
General appearance	91.75	92.69	100.07	83.96	95.69	91.27	90.51	75.80	6.09	0.05	0.71	0.25	0.03	0.07
Chewiness	71.71	69.66	76.29	76.27	78.02	69.47	81.82	63.87	6.25	0.37	0.45	0.44	0.08	0.87
Toughness	78.56	74.78	81.5	78.32	74.81	70.57	88.95	68.71	6.04	0.21	0.56	0.56	0.04	0.30
Juiciness	63.01	66.41	66.13	66.91	68.62	61.81	65.55	72.72	6.43	0.92	0.50	0.89	0.79	0.51
Pork flavor	81.95	75.60	81.22	78.49	88.97	81.33	83.77	83.10	5.52	0.81	0.27	0.29	0.23	0.76
Off-flavor	59.84	57.95	59.49	54.90	55.09	46.95	47.14	54.45	6.00	0.58	0.51	0.34	0.64	0.96
After taste pork flavor	79.99	72.76	80.70	78.11	86.88	75.55	85.32	81.29	5.65	0.65	0.30	0.21	0.09	0.69
Overall acceptability	77.29	75.27	80.43	75.82	83.46	75.74	86.85	74.89	5.85	0.71	0.41	0.49	0.09	0.80

[a]G1 = Grower1; G2 = Grower 2; F1 = Finisher 1.
[b]F2 = Finisher 2.
[c]SEM = Standard error of the mean.
[d]TRT = Treatment.
[e]L = Linear.
[f]Q = Quadratic.
[g]W = Withdrawal; W × RAC interaction, $P > 0.10$.
[h]Attribute description provided in Table 1.

dietary DDGS inclusion ($P = 0.12$). No changes were detected in chemical composition of LM in response to dietary DDGS, RAC inclusion, or DDGS withdrawal ($P > 0.10$); however, numeric increase in LM protein concentration in response to RAC inclusion were observed ($P = 0.12$).

Shear force was not affected by dietary DDGS, RAC inclusion, or DDGS withdrawal ($P > 0.10$). Dietary DDGS did not change color characteristics of the LM on day 0 (Table 3; $P > 0.10$); however, the addition of RAC resulted in decreased a* (redness; $P = 0.01$), and b* (yellowness: $P = 0.01$), which agrees with data reported by other authors. In the present study, the inclusion of RAC also decreased L* (lightness) in LM ($P = 0.04$). On day 7, RAC inclusion produced a reduction in b* and L* ($P = 0.02$ and 0.01, respectively); however, a* was not affected by RAC (P = 0.12).

The backfat fatty acid profile is presented in Table 4. The concentration of myristic acid did not change in response to increased dietary DDGS inclusion ($P = 0.14$ and 0.81 for linear and quadratic responses, respectively). The withdrawal of DDGS increased the concentration of myristic acid ($P = 0.05$). Palmitic and stearic acids concentration in backfat linearly decreased with increased concentration of dietary DDGS ($P = 0.01$ and < 0.01, respectively). Palmitic acid increased in response to DDGS withdrawal ($P < 0.01$); however, a reduction was detected for this fatty acid in response to RAC inclusion ($P = 0.02$). The withdrawal of DDGS resulted in increased stearic acid concentration ($P < 0.01$). Palmitoleic, oleic, and linoleic acids concentrations were affected by dietary treatment ($P < 0.05$). Palmitoleic showed a negative linear response to increased dietary DDGS inclusion ($P = 0.02$), and was not affected by the inclusion of RAC ($P = 0.88$). Withdrawing DDGS resulted in increased palmitoleic and reduced linoleic concentration ($P = 0.01$ and < 0.01, respectively). The inclusion of increasing concentration of dietary DDGS resulted in a linear increase in the concentration of α-linoleic acid ($P < 0.01$); similarly, the inclusion of RAC increased this fatty acid concentration in backfat ($P = 0.02$). In contrast, a reduction in α-linoleic concentration in response to DDGS withdrawal was detected ($P < 0.01$). Total saturated fatty acids concentration showed a negative linear response to increasing concentration of dietary DDGS ($P < 0.01$); similarly, the inclusion of RAC negatively affected TSFA concentration ($P = 0.03$). Increment in TSFA in response to DDGS withdrawal was detected ($P < 0.01$). Total monounsaturated fatty acid was unchanged by increasing concentration of dietary DDGS ($P = 0.60$), and increased in response to RAC inclusion ($P = 0.04$). Total polyunsaturated fatty acids concentration linearly increased in response to greater concentration of dietary DDGS ($P < 0.01$); in contrast, a reduction in TPUFA was detected in response to DDGS withdrawal ($P < 0.01$).

(Continued on next page)

Dietary DDGS did not alter the sensory characteristics of the LM (Table 5; $P > 0.10$). This is in agreement with results reported in similar studies. The inclusion of RAC resulted in increased toughness ($P = 0.04$) and a trend for increased chewiness ($P = 0.08$), which is in agreement with other studies in which the use of RAC resulted in reduced tenderness and increased chewiness. The inclusion of RAC also resulted in reduced scores for general appearance ($P = 0.03$) which is an indication of reduced uniformity in meat color. Despite the lack of treatment effect ($P = 0.65$), LM showed a tendency to have a reduced aftertaste pork flavor, and overall acceptability in response to RAC inclusion ($P = 0.09$). No effect of DDGS withdrawal was detected for any of the sensory characteristics evaluated in the present study ($P > 0.10$); however, the general appearance of the LM showed a tendency to be less uniform with DDGS withdrawal ($P = 0.07$).

Summary

The results of this investigation suggest that increasing dietary concentration of DDGS, ractopamine inclusion, or DDGS withdrawal did not affect carcass characteristics of growing-finishing pigs from the UNL Nutrition Line.

Sensory characteristics, color, and chemical composition of longissimus muscle did not change in response to increasing concentration of DDGS up to 40%, or DDGS withdrawal. The inclusion of RAC resulted in altered color characteristics of the longisimuss muscle at days 0 and 7 of retail display.

The inclusion of RAC 4 weeks before harvesting did not alleviate the changes in fatty acid profile that resulted from the inclusion of DDGS in the diet of growing-finishing pigs.

The results of the present study suggest dietary inclusion of DDGS may result in an increase in TUSFA and a decrease in TSFA in backfat of growing-finishing pigs; however, withdrawing DDGS during the last 4 weeks of the finishing period may partially reverse the changes in fatty acid profile that result from the inclusion of dietary DDGS up to 40%. The "soft pork" problems associated with changes in fatty acid profile due to dietary DDGS inclusion, may be partially resolved by withdrawing DDGS from the diet of finishing pigs 4 weeks prior harvesting.

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The Effect of Corn Distillers Dried Grains With Solubles (DDGS) on Carcass Quality, Color Stability, and Sensory Characteristics of Pork

Dietary distillers dried grain with soluble (DDGS) inclusion decreased saturated fatty acid and increased unsaturated fatty acid concentrations in fat samples from growing-finishing pigs. Concentration of dietary DDGS does not affect color, chemical composition, or sensory characteristics of pork.

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Summary

A study was conducted to evaluate the effect of feeding 0, 5, 10, or 15% distillers dried grains with solubles (DDGS) on carcass quality, color stability, and sensory characteristics of the longissimus muscle (LM) of finishing pigs. Two hundred forty pigs (61.7 lb) were assigned to 1 of 4 dietary treatments with varying concentrations of DDGS (0, 5, 10, and 15%). Live weight, hot carcass weight, and dressing percentage did not change in response to increased dietary DDGS ($P = 0.491$, 0.807, 0.316, respectively). After 7 days of retail display, yellowness changed due to DDGS inclusion ($P = 0.016$). No differences in shear force were observed ($P = 0.06$). Total polyunsaturated fatty acids increased and total saturated fatty acids decreased ($P < 0.01$, and 0.04, respectively) as dietary DDGS increased. Treatments did not differ in sensory characteristics ($P > 0.10$). The results of this investigation suggest that increasing dietary DDGS did not affect carcass quality as evaluated by color, chemical composition, and sensory characteristics of LM of finishing pigs. Increasing concentration of DDGS altered the backfat fatty acid profile of pigs by reducing

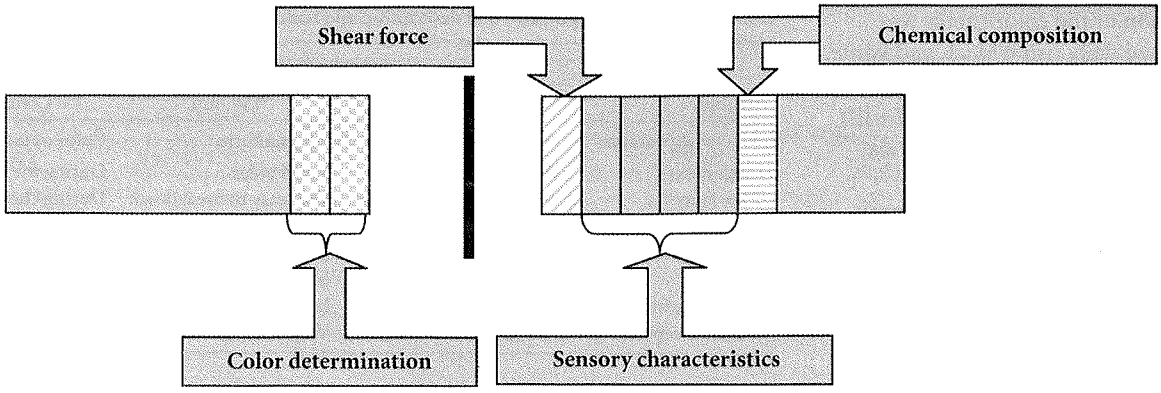

Figure 1. Longissimus muscle sections of the loins used for shear force, color determination, sensory characteristics, and chemical analysis.

saturated fatty acids and increasing unsaturated fatty acid concentration which may result in increased carcass softness.

Introduction

In recent years, an increased demand for ethanol from corn has resulted in the construction of several ethanol plants. Consequently, the amount of distillers dried grains with solubles (DDGS) available for animal feeding has also increased. Carcass quality is influenced by dietary ingredients, and evidence indicates that dietary inclusion of DDGS may result in reduced dressing percentage, as well as changes in the fatty acid profile of adipose tissue. Research evidence suggests that when dietary concentrations of DDGS greater than 20% are fed to growing-finishing pigs, the increment in unsaturated fatty acids in fat may result in increased iodine value, which may be an indication of increments in carcass softness. Additionally, changes in the fatty acid profile of carcass may result in changes in sensory characteristics such as color, flavor, and tenderness of pork.

This report is a companion article to a previous article in the *2009 Nebraska Swine Report* in which the feeding value of diets for growing-finishing pigs with increasing DDGS concentration was reported. The objective of this study was to evaluate the effects of feeding varying concentration of DDGS on carcass and sensory characteristics of pork.

Materials and Methods

Carcass Data Collection

Two hundred forty pigs weighing an average of 61.7 lb were assigned to 1 of 4 dietary treatments. Each treatment consisted of a standard diet in which a portion of dietary corn and soybean meal was replaced with 0, 5, 10, and 15% of DDGS. Diets were formulated in a SID lys basis and arranged in a 4-phase dietary growing-finishing regime. Details of the growth study are described in a companion article (*2009 Nebraska Swine Report*). At the end of the feeding phase, all pigs were transported to a commercial pork packing facility located approximately 145 miles from the University of Nebraska Swine Research Unit. Pigs were weighed before entering (live weigh; LW) and before leaving the harvesting floor (hot carcass weight; HCW). Dressing percentage (DP) was calculated using the following formula DP = ((LW / HCW) × 100). Before chilling, carcass 10th rib and LM depth were measured with a Fat-o-meater automated probe (SFK Technology AIS, Denmark), and LM area and lean percent were calculated. Iodine values were estimated using near-infrared spectroscopy at the packing plant. Carcasses were subjected to a standard spray-chilling procedure for 24 hours. Before entering the fabrication floor, two backfat samples were obtained (perpendicular to the 10th rib from the inner and outer layer), submerged in liquid nitrogen and maintained at -112°F until analyzed for fatty acid profile. Two pigs from each pen were randomly selected prior to harvesting; carcasses were identified on the chilling floor, and the bone-in loin (410 pork loin; NAMP, 1997. The Meat Buyer Guide. North American Meat Processors Association. Reston, Va.) from the right side of the carcass was collected. The collected loins were transported to the Meat Science Laboratory at the University of Nebraska for further analysis. Seven days post-mortem the loins were boned and a section of LM (412B pork loin, boneless, center-cut, eight ribs; NAMP, 1997. The Meat Buyer Guide. North American Meat Processors Association. Reston, Va.). Nine 1-inch sections (Figure 1) were obtained and used for color determination, shear force estimation, sensory characteristics evaluation, and chemical composition.

Color Determination

The two sections of the LM used for color determination were packed in Styrofoam trays, wrapped with PVC film, and maintained at 34°F under fluorescent light illumination for 7 days. Color spectrometry measurements L*, a*, and b* (representing lightness, redness, and yellowness, respectively) were obtained through the packing film on five sites on each section at the beginning (day 0) of the 7 day color experiment and daily thereafter using a Hunter Lab® Mini Scan XE plus (Model 45/0-L, Reston, Va.) handheld colorimeter. The

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calibration of the colorimeter was performed daily using black and white tiles. The change in total color (E) was calculated as [((L* at day 10 – L* at day 0)^2 + (a* at day 10 – a* at day 0)^2 + (b* at day 10 – b* at day 0)^2)^{½}; Minolta, 1998. Precise color communication-color control from perception from instrumentation. Minolta Corp., Ramsey, N.J.]. This formula was developed in order to better describe the changes in color that would occur during periods of retail display.

Warner-Bratzler Shear Force Analysis

The loin sections used for Warner-Bratzel shear force (American Meat Science Association. Research guidelines for cookery, sensory evaluation and tenderness measurements of meat. 1995) were vacuum-packed and maintained at -4°F until analysis. Before the analysis chops were allowed to thaw, cooked to an internal temperature of 158°F on a Hamilton Beach® grill (Washington, N.C.), and cooled for four hours at 35.6°F. During the cooking process temperature was monitored using thermocouples. Three cores of 0.5 in^2 from each section were removed parallel to the arrangement of the muscle fiber. Cores were sheared parallel to the muscle fiber using an Intron Universal Testing Machine (Model 55R1123, Canton, Mass.) equipped with a Warner-Bratzler shear attachment. The speed for the test was 250 mm/min.

Table 1. Attribute, magnitude, and description and scale of sensory characteristics.

Attribute	Magnitude 0 mm	Magnitude 150 mm	Comments
General appearance	Very non-uniform	Very uniform	Color of interior meat
Toughness	Very tough	Very tender	During the first bite
Chewiness	Very hard to breakdown	Very easy to breakdown	During chewing
Juiciness	Very dry	Very moist	
Pork flavor	Lacking	Intense	
Off-flavor	Lacking	Intense	
Aftertaste pork flavor	Lacking	Intense	
Overall acceptability	Very undesirable	Very desirable	

Fatty Acid Profile

Fatty acids concentration was measured in the backfat inner and outer layer of two pigs per pen. Fatty acids were extracted in hexane and methyl esters were formed. The mass ratios of fatty acids were quantified using a gas chromatograph (Hewlett-Packard, Model 5890, Farmington Hills, Mich.).

Sensory Evaluation

Chops were cooked and sensory evaluation was conducted using 70 consumer panelists, recruited from the Animal Science Department and the Department of Food Science and Technology at the University of Nebraska–Lincoln. The chops were cooked using an electric grill to an internal temperature of 158°F. Once cooked, chops were trimmed of excess fat. Samples

Table 2. Response and significance of dietary DDGS[a] inclusion on final weight and carcass characteristics of growing-finishing pigs.

Item	DDGS[a], % 0	5	10	15	SEM[b]	*P*-value Treatment	Linear	Quadratic
No. of samples	54	54	59	52				
Live weight, lb	281.1	274.3	275.4	268.4	5.40	0.491	0.163	0.903
Hot carcass weight, lb	208.4	207.6	204.9	203.5	4.02	0.807	0.344	0.938
Dressing percentage, %	74.20	74.90	73.80	74.40	0.4	0.316	0.751	0.867
Percent lean, %	51.2	52.2	52.0	52.7	0.3	0.017	0.004	0.588
10th rib BF[c], in	0.92	0.81	0.85	0.79	0.022	0.003	0.002	0.245
LMA[d], in^2	7.06	7.13	6.99	7.05	0.178	0.955	0.843	0.975
Iodine value								
Jowl	73.52	73.58	73.47	74.04	0.477	0.825	0.507	0.596
Belly	65.98	66.72	65.30	65.30	0.809	0.562	0.351	0.647
Loin	68.16	68.31	66.32	68.33	0.624	0.044	0.906	0.063
Primal cuts[e], %								
Belly	16.05	15.00	15.70	14.30	0.90	0.434	0.184	0.930
Butt	8.30	8.50	8.70	9.20	0.40	0.498	0.149	0.679
Ham	23.88	23.50	24.80	23.50	0.90	0.661	0.931	0.567
Loin	22.00	21.50	23.20	21.70	0.70	0.313	0.825	0.463
Ribs	4.80	5.00	4.80	4.80	0.20	0.944	0.874	0.724
Picnic	11.20	11.50	11.20	11.30	0.40	0.937	0.932	0.849

[a]DDGS = Corn distillers dried grains with solubles.
[b]SEM = Standard error of the mean.
[c]BF = Backfat.
[d]LMA = Longissimus muscle area.
[e]Percentage of hot carcass weight.

Table 3. Response and significance of dietary DDGS[a] inclusion on color, shear force and chemical composition of the longissumus muscle of growing-finishing pigs.

	DDGS[a], %					*P*-value		
Item	0	5	10	15	SEM[b]	Treatment	Linear	Quadratic
No. of samples	13	11	12	11				
Composition, %								
Crude protein	23.12	23.47	23.48	23.53	0.21	0.17	0.06	0.27
Moisture	68.83	70.14	69.92	70.01	0.50	0.08	0.07	0.13
Ash	1.12	1.16	1.16	1.16	0.02	0.52	0.25	0.37
Fat	6.45	4.92	5.09	5.03	0.45	0.10	0.05	0.12
Shear force, lb	7.98	9.87	9.85	9.19	0.26	0.06	0.13	0.02
Color (day 0)								
a* (redness)	18.65	18.23	18.34	18.02	0.355	0.645	0.263	0.886
b* (yellowness)	15.77	14.91	14.84	14.71	0.297	0.061	0.018	0.212
L* (lightness)	50.01	49.45	49.45	49.32	0.614	0.861	0.455	0.727
Color (day 7)								
a* (redness)	14.34	14.76	13.95	13.37	0.588	0.552	0.163	0.388
b* (yellowness)	15.55	15.34	14.99	14.82	0.189	0.016	0.004	0.895
L* (lightness)	51.50	50.09	47.65	50.44	1.759	0.588	0.475	0.229
E[c]	4.97	3.78	7.26	5.10	1.454	0.367	0.553	0.737

[a]DDGS = Corn distillers dried grains with solubles.
[b]SEM = Standard error of the mean.
[c]E = Change in color.

of 1 in^2 were obtained and maintained warm until served to the panelists. A descriptive scale was used to determine the effect of DDGS inclusion on pork quality and flavor. Panelists used an unstructured line-scale to evaluate the attributes provided in Table 1.

Statistical Analysis

Carcass characteristics, chemical composition, fatty acid profile, and sensory characteristics were analyzed as a complete randomized design using the MIXED procedure (SAS Inst., Inc., Cary, N.C.). Each pig was considered an experimental unit and pen was considered a random effect. Color data were analyzed as repeated measures in time using the MIXED procedure; the pig was considered the experimental unit and the tray was considered a random effect.

Results and Discussion

Carcass traits are shown in Table 2. A positive linear response to DDGS concentration was detected for percent lean ($P = 0.004$), which indicates that percentage lean increased as dietary DDGS increased. Contrastingly, 10th rib backfat linearly decreased with increased dietary DDGS inclusion ($P = 0.002$). No changes were detected for live weight, hot carcass weight, and longissimus muscle area (LMA; $P = 0.491$, 0.807, and 0.995, respectively). Dressing percentage did not change in response to dietary DDGS inclusion ($P = 0.316$). These results agree with those of the similar study reported in the *2008 Nebraska Swine Report*; however, other studies have shown reductions in DP as dietary DDGS concentration increased. Iodine value of jowl and belly fat did not change in response to dietary DDGS inclusion ($P > 0.10$). No changes in primal cut percentage in response to DDGS inclusion were detected ($P > 0.10$).

The results of the chemical analysis and color of LM are provided in Table 3. Moisture and ash were not affected by dietary DDGS inclusion ($P = 0.08$, and 0.52, respectively). Protein concentration showed a trend to linearly increase in response to increasing dietary DDGS inclusion ($P = 0.06$). In contrast, fat concentration showed a trend to decrease in response to increased dietary DDGS inclusion ($P = 0.05$).

Shear force did not differ among treatments ($P = 0.06$). At days 0 and 7 there was no difference among treatments for redness (a*), yellowness (b*), lightness (L*), and color change (E). These results indicate that during the 7-day experimental period, sections of loins from pigs receiving increasing dietary concentration of DDGS showed a pattern in color and change of color (E) similar to those fed the control diet (0% DDGS).

Tables 4 and 5 show the fatty acid profiles of samples of inner and outer layers of backfat. For the inner layer, myristic, palmitic, palmitoleic, and oleic were not affected by dietary DDGS concentration ($P > 0.10$). Treatments affected stearic and linoleic concentrations ($P = 0.01$ and < 0.01, respectively). A linear reduction in stearic (18:0) was detected with increased dietary DDGS concentration ($P = 0.01$). In contrast a linear increase in linoleic (18:2) was recorded with increased dietary DDGS concentration ($P < 0.01$). Treatment affected the concentration of total saturated fatty acids ($P = 0.04$). A linear reduction in total saturated fatty acids mass

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Table 4. Response and significance of dietary DDGS[a] inclusion on fatty acid profile of the inner layer of backfat of finishing pigs.

	DDGS[a], %					*P*-value		
Item	0	5	10	15	SEM[b]	Treatment	Linear	Quadratic
No. of samples	12	12	12	12				
Inner layer fatty acid, mass %								
Myristic, (14:0)	1.39	1.31	1.33	1.39	0.42	0.29	0.93	0.06
Palmitic, (16:0)	23.87	23.10	22.72	22.94	0.43	0.23	0.10	0.23
Palmitoleic, (16:1)	2.39	2.21	2.24	2.48	0.10	0.25	0.52	0.05
Stearic, (18:0)	12.63	12.53	11.69	11.16	0.35	0.01	0.01	0.45
Oleic, (18:1)	42.79	43.01	42.25	41.43	0.49	0.11	0.03	0.27
Linoleic, (18:2)	9.95	11.74	12.59	13.77	0.49	<.01	<.01	0.51
α-linolenic, (18:3)	0.40	0.43	0.44	0.47	0.012	0.09	0.01	0.82
Others	6.54	6.64	6.74	6.39	0.43	0.24	0.75	0.50
Total saturated fatty acids	37.90	36.94	35.75	35.45	0.67	0.04	<.01	0.60
Total monounsaturated fatty acids	45.18	45.23	44.67	43.91	0.51	0.29	0.04	0.53
Total polyunsaturated fatty acids	10.36	12.17	13.05	14.25	0.51	<.01	<.01	0.53

[a]DDGS = Corn distillers grains with solubles.
[b]SEM = Standard error of the mean.

Table 5. Response and significance of dietary DDGS[a] inclusion on fatty acid profile of the outer layer of backfat of finishing pigs.

	DDGS[a], %					*P*-value		
Item	0	5	10	15	SEM[b]	Treatment	Linear	Quadratic
No. of samples	12	12	12	12				
Outer layer fatty acid, mass %								
Myristic, (14:0)	1.39	1.39	1.33	1.32	0.46	0.56	0.20	0.96
Palmitic, (16:0)	22.64	21.90	21.69	21.64	0.43	0.24	0.07	0.35
Palmitoleic, (16:1)	2.40	2.28	2.42	2.36	0.11	0.76	0.97	0.78
Stearic, (18:0)	11.54	11.38	10.25	10.41	0.42	0.07	0.01	0.69
Oleic, (18:1)	43.10	42.63	42.22	41.88	0.50	0.33	0.07	0.89
Linoleic, (18:2)	11.14	12.71	13.97	14.97	0.58	<.01	<.01	0.64
α-linolenic, (18:3)	0.45	0.48	0.49	0.52	0.02	0.17	0.03	0.99
Others	7.32	7.20	7.73	6.87	0.49	0.66	0.69	0.43
Total saturated fatty acids	35.56	34.67	33.20	33.76	0.77	0.09	0.01	0.46
Total monounsaturated fatty acids	45.51	44.97	44.65	44.24	0.55	0.37	0.09	0.85
Total polyunsaturated fatty acids	11.60	13.19	14.40	15.50	0.60	<.01	<.01	0.65

[a]DDGS = Corn distillers grains with solubles.
[b]SEM = Standard error of the mean.

Table 6. Response and effect of dietary DDGS[a] inclusion on sensory characteristics of longissumus muscle of growing-finishing pigs.

	DDGS[a], %					*P*-value		
Item	0	5	10	15	SEM[b]	Treatment	Linear	Quadratic
No. of samples	12	12	12	12				
Attribute[c], mm								
General appearance	85.01	82.99	96.76	88.48	4.04	0.08	0.18	0.43
Toughness	79.02	68.04	64.87	79.42	4.31	0.05	0.98	<.01
Chewiness	74.44	64.22	61.20	69.23	4.22	0.13	0.32	0.03
Juiciness	70.31	75.86	69.30	74.67	4.12	0.60	0.72	0.98
Pork flavor	87.55	86.20	82.35	81.17	3.60	0.54	0.15	0.98
Off-flavor	47.09	52.63	52.05	57.52	4.35	0.41	0.11	0.99
Aftertaste pork flavor	84.66	79.06	75.50	79.13	3.55	0.33	0.20	0.19
Overall acceptability	87.50	82.86	77.74	82.87	4.37	0.47	0.32	0.26

[a]DDGS = Corn distillers dried grains with solubles.
[b]SEM = Standard error of the mean.
[c]Attribute description provided in Table 1.

percentage resulted from the inclusion of increasing dietary DDGS concentration ($P < 0.01$). Despite the lack of treatment effect ($P = 0.29$), total monounsaturated fatty acids mass percentage linearly decreased in response to increased dietary DDGS ($P = 0.04$). Total polyunsaturated fatty acid mass % increased linearly in response to increased concentrations of dietary DDGS ($P < 0.01$)

The outer layer backfat mass percentage of myristic, palmitic, palmitoleic, oleic, and total monounsaturated fatty exhibited no response to treatment ($P > 0.05$). However, mass percentage of stearic linearly decreased with increasing DDGS inclusion in the diets ($P = 0.01$). Mass percentage of linoleic in the outer layer of backfat exhibited treatment response ($P < 0.01$) and linearly increased with increased dietary DDGS ($P < 0.01$). Despite the lack of treatment effect ($P = 0.09$), total saturated fatty acid mass percentage, linearly decreased with increased inclusion of dietary DDGS concentration ($P = 0.01$). In contrast total polyunsaturated fatty acids exhibited a positive linear response to inclusion of increasing dietary DDGS inclusion ($P < 0.01$).

Evidence reported in the literature indicates that the inclusion of unsaturated fatty acids in the diets of growing-finishing pigs results in a reduction in the content of saturated fatty acids in adipose tissue. The results of the present study support those findings. The inclusion of DDGS in the diets of growing-finishing pigs increases the concentration of dietary unsaturated fatty acids and in consequence increases concentrations of unsaturated in the adipose tissue. Interestingly, iodine value results determined at the packing plant do not support the fatty acid results.

The effects of DDGS inclusion on sensory characteristics of longissimus muscle of finishing pigs are provided in Table 6. The inclusion of increasing dietary concentration of DDGS had minimal effects on sensory characteristics evaluated in the present study. Dietary treatment allowed ($P < 0.05$) toughness, but this effect was not consistent with increasing dietary DDGS concentration.

Conclusions

These results suggest that the inclusion of increasing levels of DDGS in diets of finishing pigs from the UNL nutrition line did not affect carcass characteristics.

Increasing dietary concentration of DDGS did not change ash or moisture concentration; however, fat concentration was reduced and protein concentration showed a tendency to increase.

Dressing percentage, color, and sensory characteristics of the LM did not exhibit changes in response to the inclusion of dietary DDGS up to 15%.

The results of this investigation suggest dietary inclusion of DDGS may result in an increase in total unsaturated fatty acid and a decrease in total saturated fatty acid concentrations.

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Utilizing Dried Distillers Grains With Solubles and Phytase to Alleviate Phosphorus Costs in Finishing Swine

Growth performance of growing-finishing pigs was maintained when calcium-phosphates were reduced or replaced with dried distillers grains, phytase, or both.

Justin W. Bundy
Phillip S. Miller
Roman Moreno
Thomas E. Burkey
Erin Hinkle
Huyen Tran[1]

Summary

A total of 24 barrows (86.7 lb) were used over a 12-week experiment to evaluate the effects of utilizing dried distillers grains with solubles (DDGS) and phytase as alternatives to inorganic phosphorus (P) sources. Pigs were assigned to 1 of 4 dietary treatments. Treatments consisted of a common corn and soybean-meal fortified diet (CSB), a diet similar to CSB with phytase added in place of dicalcium-phosphate, a corn and soybean-meal diet with 20% DDGS, and a diet with 20% DDGS and phytase added in place of dicalcium-phosphate. Treatment did not affect ($P > 0.10$) pig performance for any of the growth criteria measured. Utilizing DDGS and phytase together helped to numerically reduce the price per ton of feed compared to the other treatments in the experiment. The results of this experiment suggest that alternatives such as DDGS and phytase can be used to manipulate the necessary inclusion rate of calcium-phosphates needed in the growing-finishing phase of swine production.

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Introduction

Previously (Fall 2008), calcium-phosphates commonly used in swine diets dramatically increased in price. Phosphorus supplementation can be one of the most expensive components in diets formulated for swine. Because feed contributes up to 70% of the total cost of production, methods to lower feed costs without affecting performance can be helpful in lowering operating costs. The objective of this experiment was to evaluate the effects of DDGS and phytase, separately and together, as alternatives that could reduce the amount of calcium-phosphates needed to meet phosphorus requirements.

Procedures

Animals and Facilities

Animal use and procedures for the experiment were approved by the Institutional Animal Care and Use Committee of the University of Nebraska–Lincoln. The 12-week study used 24 crossbred barrows (initial BW = 86.7 lb). Animals were individually penned in an environmentally-controlled room with ad libitum access to feed and water. Barrows were randomly assigned to 1 of 4 dietary treatments (six pigs/treatment) for the duration of the experiment.

Dietary Treatments

The 4 dietary treatments included: a traditional corn-soybean meal diet (CSB); a CSB with 1,000 FTU of a commercially available phytase (*E. coli derived*) and no supplemental inorganic phosphorus (P); a CSB with 20% DDGS inclusion; and a CSB with 20% DDGS inclusion 1,000 FTU of phytase and no supplemental inorganic P (Tables 1, 2, and 3). Pigs were fed in 3 dietary phases (Phase 1, 90 to 130 lb; Phase 2, 130 to 180 lb; Phase 3, 180 to 260 lb). Diets were formulated to meet or exceed nutrient requirements based on NRC (1998) values, except for P which was formulated to be adequate in available P for the CSB and 20% DDGS without phytase

Table 1. Ingredient and calculated composition of the grower diet (90 to 125 lb), as-fed basis.

	Treatment			
DDGS[a], %	**0**	**0**	**20**	**20**
Phytase[b], FTU[c]	**0**	**1,000**	**0**	**1,000**
Ingredient, %				
Corn	78.71	79.01	62.80	62.95
Soybean meal-47.5% CP[d]	18.76	18.73	14.79	14.78
DDGS	0.00	0.00	20.00	20.00
Phytase	0.00	0.05	0.00	0.05
Dicalcium phosphate	0.86	0.00	0.49	0.00
Limestone	0.90	1.43	1.14	1.44
Salt	0.30	0.30	0.30	0.30
Vitamin premix[e]	0.20	0.20	0.20	0.20
Trace mineral premix[f]	0.15	0.15	0.15	0.15
Antibiotic[g]	0.03	0.03	0.03	0.03
L-Lysine•HCl	0.10	0.10	0.10	0.10
Calculated analysis, %				
Total lysine, %	0.85	0.85	0.85	0.85
CP, %	15.54	15.55	17.99	17.99
Calcium, %	0.60	0.60	0.60	0.60
Phosphorus, %	0.51	0.35	0.49	0.40
Available phosphorus, %	0.22	0.06	0.22	0.13
ME[h], kcal[i]/lb	1,512	1,516	1,558	1,560
Calculated cost/ton				
August 27, 2008, $/ton	250.9	244.4	238.8	235.9

[a]DDGS = Corn dried distillers grains with solubles.
[b]Optiphos®, JBS United (Sheridan, Ind.).
[c]FTU = phytase units.
[d]CP = Crude protein.
[e]Supplied per kilogram of diet at 0.2% inclusion: vitamin A as retinyl acetate, 4,400 IU; cholecalciferol, 440 IU; α-tocopherol acetate, 24 IU; menadione sodium bisulfite, 3.5 mg; riboflavin 8.8 mg; d-pantothenic acid, 17.6 mg; niacin, 26.4 mg; vitamin B_{12}, 26.4 mg.
[f]Supplied per kilogram of diet at 0.15% inclusion: Zn (as $ZnSO_4$), 128 mg; Fe (as $FeSO_4 \bullet H_2O$), 128 mg; Mn (as MnO), 30 mg; Cu ($CuSO_4 \bullet 5\ H_2O$), 10.5 mg; I (as $Ca(IO_3) \bullet H_2O$), 0.26 mg; Se (as Na_2SeO_3), 0.26 mg.
[g]Tylan®, Elanco Animal Health (Greenfield, Ind.).
[h]ME = Metabolizable energy.
[i]kcal = kilocalories.

Table 2. Ingredient and calculated composition of finisher 1 diets (125 to 185 lb), as-fed basis.

	Treatment			
DDGS[a], %	**0**	**0**	**20**	**20**
Phytase[b], FTU[c]	**0**	**1,000**	**0**	**1,000**
Ingredient, %				
Corn	84.30	84.55	68.40	68.49
Soybean meal-47.5% CP[d]	13.31	13.29	9.34	9.34
DDGS	0.00	0.00	20.00	20.00
Phytase	0.00	0.05	0.00	0.05
Dicalcium phosphate	0.73	0.00	0.37	0.00
Limestone	0.89	1.34	1.11	1.35
Salt	0.30	0.30	0.30	0.30
Vitamin premix[e]	0.20	0.20	0.20	0.20
Trace mineral premix[f]	0.15	0.15	0.15	0.15
Antibiotic[g]	0.03	0.03	0.03	0.03
L-Lysine•HCl	0.10	0.10	0.10	0.10
Calculated analysis, %				
Total lysine, %	0.70	0.70	0.70	0.70
CP, %	13.41	13.42	15.86	15.87
Calcium, %	0.55	0.55	0.55	0.55
Phosphorus, %	0.46	0.33	0.45	0.38
Available phosphorus, %	0.19	0.05	0.19	0.12
ME[h], kcal[i]/lb	1,515	1,519	1,561	1,562
Calculated cost/ton				
August 27, 2008, $/ton	245.1	239.8	233.0	231.3

[a]DDGS = Corn dried distillers grains with solubles.
[b]Optiphos®, JBS United (Sheridan, Ind.).
[c]FTU = phytase units.
[d]CP = Crude protein.
[e]Supplied per kilogram of diet at 0.2% inclusion: vitamin A as retinyl acetate, 4,400 IU; cholecalciferol, 440 IU; α-tocopherol acetate, 24 IU; menadione sodium bisulfite, 3.5 mg; riboflavin 8.8 mg; d-pantothenic acid, 17.6 mg; niacin, 26.4 mg; vitamin B_{12}, 26.4 mg.
[f]Supplied per kilogram of diet at 0.15% inclusion: Zn (as $ZnSO_4$), 128 mg; Fe (as $FeSO_4 \bullet H_2O$), 128 mg; Mn (as MnO), 30 mg; Cu ($CuSO_4 \bullet 5\ H_2O$), 10.5 mg; I (as $Ca(IO_3) \bullet H_2O$), 0.26 mg; Se (as Na_2SeO_3), 0.26 mg.
[g]Tylan®, Elanco Animal Health (Greenfield, Ind).
[h]ME = Metabolizable energy.
[i]kcal = kilocalories.

Table 3. **Ingredient and calculated composition of finisher 2 diets (185 to 260 lb), as-fed basis.**

	Treatment			
DDGS[a], %	**0**	**0**	**20**	**20**
Phytase[b], FTU[c]	**0**	**1,000**	**0**	**1,000**
Ingredient, %				
Corn	89.89	90.09	73.98	74.03
Soybean meal-47.5%CP[d]	7.86	7.85	3.90	3.89
DDGS	0.00	0.00	20.00	20.00
Phytase	0.00	0.05	0.00	0.05
Dicalcium phosphate	0.61	0.00	0.24	0.00
Limestone	0.87	1.24	1.11	1.26
Salt	0.30	0.30	0.30	0.30
Vitamin premix[e]	0.20	0.20	0.20	0.20
Trace mineral premix[f]	0.15	0.15	0.15	0.15
Antibiotic[g]	0.03	0.03	0.03	0.03
L-Lysine•HCl	0.10	0.10	0.10	0.10
Calculated analysis, %				
Total lysine, %	0.55	0.55	0.55	0.55
CP, %	11.29	11.30	13.74	13.74
Calcium, %	0.50	0.50	0.50	0.50
Phosphorus, %	0.42	0.31	0.40	0.36
Available phosphorus, %	0.16	0.05	0.16	0.12
ME[h], kcal[i]/lb	1,518	1,521	1,564	1,565
Calculated cost/ton				
August 27, 2008, $/ton	239.2	235.2	227.1	226.8

[a]DDGS = Corn dried distillers grains with solubles.
[b]Optiphos®, JBS United (Sheridan, Ind.).
[c]FTU = phytase units.
[d]CP = Crude protein.
[e]Supplied per kilogram of diet at 0.2% inclusion: vitamin A as retinyl acetate, 4,400 IU; cholecalciferol, 440 IU; α-tocopherol acetate, 24 IU; menadione sodium bisulfite, 3.5 mg; riboflavin 8.8 mg; d-pantothenic acid, 17.6 mg; niacin, 26.4 mg; vitamin B_{12}, 26.4 mg.
[f]Supplied per kilogram of diet at 0.15% inclusion: Zn (as $ZnSO_4$), 128 mg; Fe (as $FeSO_4•H_2O$), 128 mg; Mn (as MnO), 30 mg; Cu ($CuSO_4•5\ H_2O$), 10.5 mg; I (as $Ca(IO_3)•H_2O$), 0.26 mg; Se (as Na_2SeO_3), 0.26 mg.
[g]Tylan®, Elanco Animal Health (Greenfield, Ind.).
[h]ME = Metabolizable energy.
[i]kcal = kilocalories.

Table 4. **Growth performance data from the dietary treatments.**

	Treatment					
DDGS, %	0	0	20	20		
Phytase, FTU	0	1,000	0	1,000		
Number of pigs	6	6	6	6	SEM[a]	*P*-value treatment
Grower (90 to 125 lb)						
ADG[c], lb	2.11	1.73	1.87	2.04	0.11	0.13
ADFI[d], lb	5.29	4.75	5.02	5.51	0.23	0.13
G:F[e], lb/lb	0.40	0.36	0.37	0.37	0.02	0.65
Finisher 1 (125 to 185 lb)						
ADG, lb	2.21	2.24	2.23	2.19	0.16	0.99
ADFI, lb	6.66	6.68	6.68	6.99	0.42	0.94
G:F, lb/lb	0.33	0.34	0.34	0.32	0.02	0.90
Finisher 2 (185 to 260 lb)						
ADG, lb	2.08	2.25	1.93	2.01	0.22	0.78
ADFI, lb	7.93	7.91	7.72	7.44	0.55	0.91
G:F, lb/lb	0.26	0.28	0.25	0.27	0.02	0.63
Overall (90 to 260 lb)						
ADG, lb	2.13	2.16	2.02	2.08	0.15	0.96
ADFI, lb	6.89	6.76	6.74	6.84	0.36	0.99
G:F, lb/lb	0.31	0.31	0.30	0.31	0.02	0.95

[a]SEM = Standard error of the mean.
[b]BW = Body weight.
[c]ADG = Average daily gain.
[d]ADFI = Average daily feed intake.
[e]G:F = Gain to feed ratio.

treatments (Table 1). Inclusion of 20% DDGS allowed for a reduction in the amount of dicalcium-phosphate needed to meet P requirements. Based on claims by the manufacturers of the phytase product used, 0.05% added phytase liberates up to 0.20% available P. Therefore, the available P shown in Table 1 represents only the available P estimates before phytase liberation of additional available P. Phytase inclusion remained the same for all 3 phases to supply available P at or above the requirements.

Data Collection

Pigs and feeders were weighed at the beginning and end of each phase. Estimated feed disappearance was calculated by the difference between feed added and the amount of feed remaining in the feeder. Body weight gain was estimated using the difference of the observed weight at the end and the beginning weight of each phase. Based on the feed disappearance and body weight gain data, average daily gain (ADG), average daily feed intake (ADFI), and feed efficiency (G:F) were estimated.

Statistical Analysis

Data were analyzed using the MIXED procedure (SAS Inst. Inc. Cary, N.C.). Each animal was considered the experimental unit. The model was a completely randomized design.

Results and Discussion

The growth performance data are provided in Table 4. During the grower period (90 to 125 lb) treatment did not affect ADG, ADFI, or G:F ($P > 0.10$). Numerically, pigs fed the diet with phytase inclusion and no DDGS exhibited the lowest for ADG and ADFI (1.73 and 4.75 lb, respectively). This difference (although only numeric) could be due to the P requirement of barrows during this phase of growth (90 to 125 lb) compared to the amount of P that the phytase needed to liberate in conjunction with decreased feed intake. During the grower period, the

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combination of DDGS and phytase resulted in the greatest reduction of price per ton ($15) compared to the traditional diet without affecting animal performance.

During the second and third phase feeding periods of the experiment (125 to 185 and 185 to 260 lb, respectively) there were no differences in ADG, ADFI, or G:F ($P > 0.10$). Numerical differences between treatments were small, which would indicate that phytase and DDGS inclusion in diets separately or together can reduce or eliminate the need of calcium-phosphates during the last stages of production.

There were no differences observed among the treatments during the three phases of the experiment or the growing-finishing period overall (90 to 260 lb) for the animal performance characteristics measured. The costs per ton of each dietary treatment indicates that the addition of DDGS and phytase separately or together can reduce feed cost based on August 27, 2008 prices. The greatest reduction in feed cost (up to $15/ton) was observed when both DDGS and phytase were used together. It is important to note that DDGS inclusion in diets can also reduce the amount of other ingredients which also contribute to the reduction in feed cost. These results are in agreement with concurrent research conducted at other universities which have concluded that DDGS along with phytase inclusions in nursery diets can alleviate any need for supplementing diets with calcium-phosphates. The results of this experiment indicate that proper formulation of diets with phytase and DDGS can reduce or alleviate dependence on traditional phosphorus sources in swine grower-finisher diets.

Conclusions

Overall, animal performance did not differ when alternative methods of P supplementation were used in growing-finish pigs (90 to 260 lb). These results suggest that expensive sources of P can be omitted from diets in order to decrease feed costs without altering animal growth parameters.

[1]Justin W. Bundy, graduate student; Phillip S. Miller, professor; Roman Moreno, graduate student and a research technologist; Thomas Burkey, assistant professor; Erin Hinkle, graduate student; and Huyen Tran, graduate student, Animal Science Department, University of Nebraska, Lincoln, Neb.

Effects of Distillers Dried Grains With Solubles and Lactose on Growth Performance of Nursery Pigs

There was no interaction between lactose and DDGS, but lactose can be incorporated in nursery diet containing DDGS and maintain growth performance.

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Justin W. Bundy
Erin Hinkle
Thomas E. Burkey
Phillip S. Miller[1]

Summary

A 4-week feeding experiment was conducted to evaluate effects of distillers dried grains with solubles (DDGS) and lactose on growth performance of nursery pigs. Ninety-six pigs (age, 23 ± 2 days; initial body weight, 14.15 ± 0.11 lb) were randomly allotted into each of 16 pens by gender, ancestry, and weight (6 pigs/pen; 4 pens/treatment). In phase 1 (weeks 1 and 2), pigs were fed 1 of the 4 treatments: A) control (no DDGS and lactose), B) 15% DDGS, C) 20% lactose, D) 15% DDGS + 20% lactose. In phase 2 (weeks 3 and 4), all pigs were fed a common diet containing 15% DDGS and 10% lactose. Diets were formulated to contain 1.47 and 1.42% true ileal digestible Lys in phase 1 and 2, respectively. Pigs receiving DDGS in phase 1 (Treatments B and D) had greater ADG and ADFI ($P = 0.05$ and 0.004, respectively) during phase 2 compared to non-DDGS fed pigs in phase 1 (Treatment A and C); however, no DDGS effects were observed on ADG, ADFI, and G:F in phase 1. Pigs receiving lactose in Phase 1 (Treatments C and D) had 21% greater ADG, 12% greater G:F ($P = 0.01$), and a trend of increased ADFI ($P = 0.07$) during phase 1, but decreased ADG during phase 2 ($P = 0.09$) compared to pigs that did not receive lactose in phase 1 (Treatments A and B). In conclusion, although there was no interaction between DDGS and lactose in any phase of this experiment, it appeared that lactose can be incorporated in nursery diets containing DDGS and have growth performance maintained.

Introduction

Distillers dried grains with solubles (DDGS) has been included in diets for growing-finishing pigs and sows, but very limited levels for weanling pigs have been used due to

Table 1. Composition of ingredients and calculated analysis, as-fed basis (%).

	Phase 1[a]				Phase 2
Treatment	A	B	C	D	Common diet
DDGS, %	0	15	0	15	15
Lactose, %	0	0	20	20	10
Ingredients, %					
Corn	61.5	47.0	37.0	22.5	36.1
DDGS	0	15	0	15	15
Soybean meal, 46.5% CP	20.5	20.5	20.5	20.5	20.5
Spray-dried porcine plasma	5	5	5	5	2.5
Select menhaden fish meal	6	6	6	6	7.5
DairyLac 80	0	0	25	25	12.5
Dicalcium phosphate, 18.5%P	1.2	0.775	0.4	0	0
Limestone	0.325	0.500	0.450	0.600	0.475
Salt	0.3	0.3	0.3	0.3	0.3
Zinc oxide	0.3	0.3	0.3	0.3	0.3
Vitamin premix[b]	0.25	0.25	0.25	0.25	0.25
Trace mineral premix[c]	0.15	0.15	0.15	0.15	0.15
L-Lysine•HCl	0.285	0.220	0.350	0.285	0.325
DL-Methionine	0.120	0.030	0.185	0.095	0.070
L-Threonine	0.095	0.015	0.138	0.060	0.070
Mecadox 2.5	1	1	1	1	1
Corn oil	3	3	3	3	
Calculated analysis					
CP[d],%	22.8	25.6	21.7	24.4	24.2
Total Lys, %	1.60	1.60	1.60	1.60	1.56
Tid[e] Lys, %	1.47	1.47	1.47	1.47	1.42
Ca, %	0.85	0.85	0.85	0.85	0.82
P, %	0.79	0.78	0.73	0.72	0.69
Available P, %	0.51	0.51	0.51	0.51	0.45
ME[f], kcal[g]/lb	1,531	1,566	1,529	1,564	1,567

[a]Phase 1 diets included: control (A); corn distillers dried grains with soluble (DDGS; B); lactose (C); DDGS plus lactose (D).
[b]Vitamin premix containing: vitamin A as retinyl acetate, 5,500 IU; vitamin D_3 as cholecalciferol, 550 IU; vitamin E as alpha-tocopherol acetate, 30 IU; vitamin K as menadione dimethylpyrimidinol bisulfide, 4.4 mg/kg; niacin, 33 mg/kg, pantothenic acid as d-Calcium pantothenate, 22.05 mg/kg; riboflavin, 11 mg/kg; vitamin B12 as cyanocobalamin, 33 mg/kg.
[c]Trace mineral premix containing: copper (as $CuSO_4•5H_2O$), 10 mg/kg; iodine (as $Ca(IO_3)•H_2O$), 0.25 mg/kg; Iron ($FeSO_4•2H_2O$), 125 mg/kg; manganese (MnO), 15 mg/kg; Selenium (Na_2SeO_3), 0.3 mg/kg; Zinc ($ZnSO_4•H_2O$), 125 mg/kg
[d]Crude protein.
[e]True ileal digestible.
[f]Metabolizable energy.
[g]kcal=Kilocalories (1,000 cal)

high percentage of insoluble fiber, low starch, and variable amino acid profiles. Publications evaluating effects of feeding DDGS on growth performance of nursery pigs are not numerous and inconsistent results were reported among studies. Some studies have shown that feeding DDGS from 5 to 25% for nursery pigs after 2 or 3 weeks postweaning did not affect weight gain, feed intake, and feed efficiency. In contrast, early introduction of DDGS at high concentration for weanling pigs could decrease average daily gain and feed intake. Because of the expansion of ethanol production concomitant with the increasing DDGS availability, investigating the use of DDGS for nursery pigs is warranted.

Inclusion of dried whey in nursery diets improves growth performance of nursery pigs. It appears that lactose and lacalbumin components in dried whey are responsible for the beneficial effects of dried whey on weanling-pig performance. Also, it has been shown that deproteinized whey can replace the lactose fraction provided by dried whey without affecting growth performance.

The effects of incorporating DDGS with other supplementary ingredients such as milk products on nursery pig performance have received little attention. We hypothesized that supplementation of the lactose fraction of deproteinized whey can compensate for the deficiency of soluble fiber and starch in DDGS diets and consequently improve growth performance of pigs fed DDGS. Therefore, this study was conducted to determine the effects of lactose, DDGS, and their interaction on growth performance of nursery pigs.

Materials and Methods

Animals and Experimental Design

The experimental protocol was reviewed and approved by the Institutional Animal Care and Use Committee of the University of Nebraska–Lincoln. Ninety-six pigs (48 barrows and 48 gilts) weaned at 23 ± 2 days were randomly allotted in 16 pens by ancestry, initial body weight, and sex (6 pigs/pen). The initial average pig BW was 14.15 ± 0.11 lb. There were four pen replications per treatment with three barrows and three gilts per pen. Pigs were housed in the temperature-controlled room and each pen had a single nipple waterer and a single self-feeder for ad libitum access to water and feed. The study duration was 4 weeks and divided into phase 1 (week 1 to 2) and phase 2 (week 3 to 4).

Dietary Treatments

Dietary treatments were arranged as a 2 × 2 factorial containing the following: A) control (no DDGS and lactose); B) 20% lactose; C) 15% DDGS; and D) 15% DDGS + 20% lactose. The ingredient compositions and calculated analysis of experimental diets are presented in Table 1. Diets in phase 1 and phase 2 were formulated to contain 1.47% and 1.42% true ileal digestible Lys, respectively. Total lysine was 1.60% for all diets in phase 1 and 1.56% for a common diet fed to all treatment groups in phase 2. This common diet was designed to contain 15% DDGS and 10% lactose. Dairylac 80 (International Ingredient Corpora-

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tion, IIC, Mo.), containing 3.2% CP, and 0.06% Lys (analyzed composition) and 80% lactose, was the only source of lactose in this experiment. This product was granular and nonhygroscopic, produced from sweet and dried whey soluble (Cromwell et al. 2008). All amino acids, vitamins, and minerals were formulated to meet or exceed the requirement identified by NRC (1998).

Data and Sample Collection

Individual pig BW and feed disappearance were recorded at the beginning of the experiment and weekly thereafter to calculate average daily gain (ADG), average daily feed intake (ADFI), and ADG:ADFI (G:F ratio).

Statistical Analysis

Growth data were analyzed as a completely randomized design (2 × 2 factorial) using the MIXED procedure of SAS. Each pen was considered an experimental unit. Model included the main effects of DDGS, lactose, and their interaction. Pen was a random effect.

Results and Discussion

Pig BW and growth performance are shown in Table 2. Initial and final BW were 14.15 ± 0.11 and 37.29 ± 0.75 lb, respectively. There was no interaction of DDGS and lactose on pig BW at any phase of this experiment. Except for a lactose effect observed on BW at the end of phase 1 ($P = 0.03$), there was no lactose effect on BW. No DDGS effect was recorded in weeks 1 and 2. However, compared to pigs that did not receive DDGS in phase 1 (week 1 to 2; Treatments A and C), pigs receiving DDGS (Treatments B and D) tended to exhibit increased BW at weeks 3 and 4 ($P = 0.08$ and 0.1, respectively) after being introduced to a common diet containing 15% DDGS and 10% lactose.

There was no interaction between DDGS and lactose on ADG, ADFI, and G:F ratio at any phase of the experiment. However, there were tendencies of lactose to increase ADG at week 1 ($P = 0.1$) and 2 ($P = 0.06$), and a significant lactose effect on G:F ratio ($P = 0.04$) at week 2. During phase 1 (week 1 to 2), ADG and G:F were greater ($P = 0.01$; 21% and 12% greater, respectively) in pigs fed lactose compared to pigs fed no lactose. In addition, pigs fed lactose tended ($P = 0.07$) to have greater ADFI compared to pigs fed no lactose in phase 1. These results are consistent with previous studies that reported feeding lactose during the early phases of the weanling period can increase ADG and G:F. In contrast, DDGS did not affect ADG, ADFI, and G:F in phase 1 (week 1 to 2) of the experiment. Our results agreed with previous studies that showed feeding DDGS immediately after weaning did not affect growth performance and feed efficiency of nursery pigs. However, these results were not consistent with another publication that reported reduced growth performance in pigs introduced to DDGS early in the nursery phase.

There were DDGS effects on ADG and ADFI at week 3 ($P = 0.01$)

Table 2 Effects of feeding DDGS and lactose on growth performance

Treatment[a]	A	B	C	D				
DDGS, %	0	15	0	15				
Lactose, %	0	0	20	20				
						P- value		
					SEM	P_D[c]	P_L[d]	P_{Dxl}[e]
BW, lb								
Week 0	14.19	14.12	14.17	14.08	0.11	0.49	0.79	0.86
Week 1	16.13	16.15	16.30	16.63	0.20	0.39	0.15	0.54
Week 2	19.62	19.62	20.77	20.75	0.44	1.00	**0.03**	0.98
Week 3	26.97	28.16	27.68	28.36	0.48	**0.08**	0.37	0.60
Week 4	36.30	38.24	37.00	37.62	0.75	**0.10**	0.96	0.38
Phase1 (week 1)								
ADG, lb	0.28	0.29	0.31	0.36	0.03	0.19	**0.10**	0.46
ADFI, lb	0.40	0.42	0.44	0.45	0.02	0.51	0.16	0.80
G:F, lb/lb	0.70	0.69	0.69	0.82	0.04	0.18	0.18	0.15
Phase 1 (week 2)								
ADG, lb	0.50	0.49	0.63	0.59	0.06	0.64	**0.06**	0.74
ADFI, lb	0.76	0.77	0.84	0.82	0.04	0.89	0.21	0.72
G:F, lb/lb	0.65	0.63	0.75	0.71	0.04	0.50	**0.04**	0.86
Phase 2 (week 3)								
ADG, lb	1.05	1.22	0.99	1.09	0.04	**0.01**	**0.05**	0.43
ADFI, lb	1.37	1.51	1.34	1.46	0.04	**0.01**	0.36	0.75
G:F, lb/lb	0.77	0.81	0.74	0.75	0.03	0.45	0.17	0.59
Phase 2 (week 4)								
ADG, lb	1.33	1.44	1.33	1.32	0.06	0.40	0.32	0.34
ADFI, lb	1.98	2.14	1.93	2.07	0.05	**0.02**	0.26	0.86
G:F, lb/lb	0.67	0.67	0.69	0.64	0.02	0.12	0.64	0.15
Phase 1 (week 1, 2)								
ADG, lb	0.39	0.39	0.47	0.48	0.03	0.85	**0.01**	0.98
ADFI, lb	0.58	0.60	0.64	0.63	0.02	0.83	**0.07**	0.64
G:F, lb/lb	0.67	0.65	0.73	0.75	0.03	0.91	**0.01**	0.58
Phase 2 (week 3, 4)								
ADG, lb	1.19	1.33	1.16	1.21	0.04	**0.05**	**0.09**	0.30
ADFI, lb	1.67	1.82	1.63	1.76	0.04	**0.004**	0.22	0.77
G:F, lb/lb	0.71	0.73	0.71	0.68	0.02	0.81	0.21	0.26
Overall (week 1 to 4)								
ADG, lb	0.79	0.86	0.81	0.84	0.02	**0.07**	0.92	0.37
ADFI, lb	1.13	1.21	1.14	1.20	0.03	**0.02**	0.94	0.68
G:F, lb/lb	0.70	0.71	0.72	0.70	0.01	0.84	0.80	0.34

[a]Phase 1 (weeks 1 and 2) dietary treatments included control (A); corn distillers dried grains with soluble (DDGS; B); lactose (C); DDGS plus lactose (D). All pigs were fed a common diet in phase 2 (weeks 3 and 4; 15% DDGS, 10% lactose).
[b]Standard Error of the Mean.
[c]Main effect of DDGS.
[d]Main effect of lactose.
[e]DDGS × lactose interaction.

and ADFI at week 4 ($P = 0.02$). During phase 2 (week 3 to 4), greater ADG ($P = 0.05$) and ADFI ($P = 0.004$) were observed in pigs consuming a diet containing DDGS (Treatments B and D) compared to pigs that did not receive DDGS in phase 1 (Treatments A and C); however, G:F ratio was not affected by DDGS. There was a lactose effect on ADG in week 3 ($P = 0.05$) and a trend of lactose effect on ADG during phase 2 ($P = 0.09$); pigs fed lactose in phase 1 (Treatments C and D) had lower ADG compared to pigs that did not receive lactose in phase 1 (Treatments A and B). These observations reinforced the traditional lactose effects on improving pig growth performance during the early postweaning period. Also, these results indicate that lactose can be added to the DDGS-containing diets and maintain growth performance.

For the overall experimental period (week 1 to 4), greater ADFI ($P = 0.02$) and ADG ($P = 0.07$) were observed in pigs fed DDGS (Treatments B and D) compared to pigs that did not receive DDGS in phase 1 (Treatments A and C); however, no effects of DDGS on G:F ratio. In addition, there were no lactose and lactose × DDGS effects on ADG, ADFI, and G:F overall.

In summary, the following observations were made: 1) pigs receiving DDGS in phase 1 (Treatments B and D) had greater ADG and ADFI ($P = 0.05$ and 0.004, respectively) during phase 2 compared to non-DDGS fed pigs in phase 1 (Treatment A and C); 2) pigs receiving lactose in phase 1 (Treatments C and D) had greater ADG, G:F ($P = 0.01$), and ADFI ($P = 0.07$) during phase 1, but decreased ADG during phase 2 ($P = 0.09$) compared to pigs that did not receive lactose in phase 1 (Treatments A and B).

Conclusions

In conclusion, there was no interaction between DDGS and lactose on growth performance of nursery pigs in any phase of this experiment. However, the inclusion of lactose in diets containing DDGS did have positive effects on improving growth performance of nursery pigs. Additional research needs to be conducted to determine level of lactose that should be incorporated with DDGS in diets to maximize pig performance and health.

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Lysine Requirement for Barrows Fed Ractopamine

Barrows fed ractopamine during the finishing phase require 0.76% total lysine in order to maximize growth performance.

Justin W. Bundy
Phillip S. Miller
Roman Moreno
Thomas E. Burkey
Erin Hinkle
Huyen Tran[1]

Summary

A total of 36 individually penned barrows (initial weight = 176 lb) were used in an experiment conducted to determine the lysine requirement for barrows fed ractopamine during the last 85 lb of the finishing phase. There were six dietary treatments that were corn-soybean meal-based with additional crystalline amino acid supplementation. Pigs were assigned to one of six dietary treatments for the duration of the experiment. Treatments included lysine concentrations ranging from 0.5 to 1.3% total lysine. Dietary treatment significantly affected average daily gain and feed efficiency. The total lysine requirement was estimated to be 0.76%. These results indicate that the outcome of ractopamine utilization can be influenced by amino acid concentration of the diet in the finishing phase.

Introduction

Ractopamine is currently used in the commercial swine industry to increase growth performance during late finishing. It is stated by the current manufacturer of ractopamine (Elanco) that the dietary crude protein concentration of the diet should be ≥ 16%. However, there is limited information available concerning the amino acid requirements for pigs fed diets containing ractopamine. In order to maximize performance during the finishing phase of production, it is important to better understand specific amino acid requirements. An improved understanding of amino acid nutrition as it pertains to ractopamine use can allow for optimal pig performance. The objective of this experiment was to determine the lysine requirement for barrows fed ractopamine during the last finisher phase.

Procedures

Animals and Facilities

Animal use and procedures for the experiment were approved by the Intitutional Animal Care and Use Committee of the University of Nebraska–Lincoln. A total of 36 crossbred barrows (initial BW = 176 lb) were individually penned in an

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environmentally controlled room. Automatic waterers and feeders were located in each pen. Pigs were allowed ad libitum access to feed and water throughout the entire experiment.

Dietary Treatments

Each pig was randomly assigned to one of six dietary treatments. The six isocaloric dietary treatments included: a control diet without ractopamine (RAC) formulated to contain 0.7% total lysine; the control diet with the inclusion of 10 ppm RAC and 0.7% total lysine; and four additional diets with 10 ppm RAC and differing concentrations of total lysine for a total of six dietary treatments (0.5, 0.9, 1.1, and 1.3% total lysine, respectively). All dietary treatments were corn-soybean meal-based. Other amino acids were maintained at a concentration at least +10% of the true ileal digestible amino acid to lysine ratios suggested by NRC (1998; Table 1).

Data Collection

Pigs and feeders were weighed at the beginning and end of each week. Data collected included feed disappearance and weight gain in order to calculate average daily feed intake (ADFI), average daily gain (ADG), and feed efficiency (G:F). Ultrasound measurements were taken weekly to determine backfat thickness (BF) and longissimus muscle area (LMA).

Statistical Analysis

Data were analyzed as a completely randomized design using the MIXED procedure (SAS Inst. Inc. Cary, N.C.). Each animal was considered the experimental unit. To determine the optimal concentration of total lysine in the diet, broken line, nonlinear regression was used.

Results and Discussion

The growth performance results of the barrows are shown in Table 2. Dietary lysine concentration affected ADG ($P < 0.05$). The lowest ADG

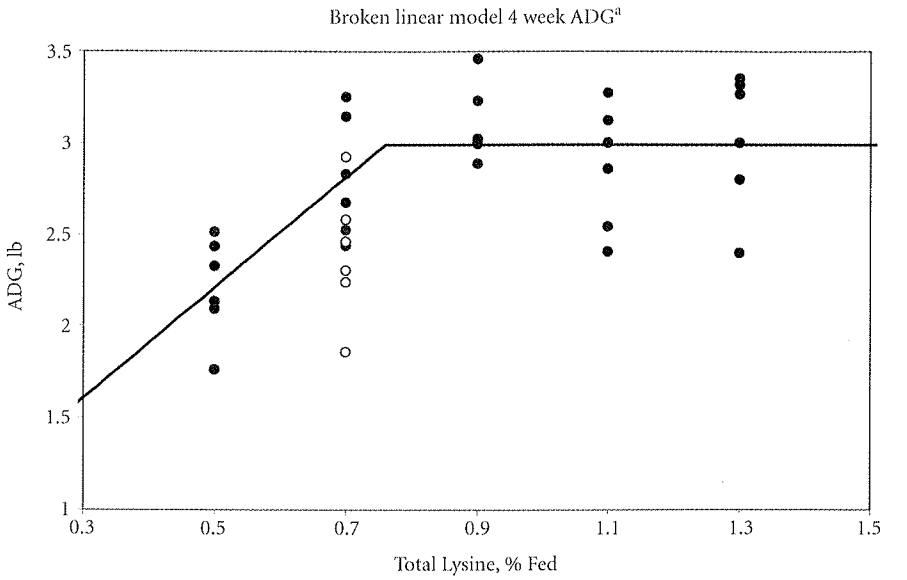

Figure 1. Observed and predicted values from four-week response data ($R^2 = 0.515$). [a]ADG = average daily gain. Black dot (●) represents ADG response for pigs fed diets containing 10 ppm ractopamine at varied total lysine %. White dot (○) represents ADG response for pigs fed diets without ractopamine and 0.7% total lysine. Solid line (—) represents predicted ADG response for pigs fed 10 ppm ractopamine at varied total lysine %. Prediction equation: If total lysine % < 0.76 then ADG = 2.99 + (−3.01 × (0.76 − total lysine %); If total lysine % ≥ 0.76 then ADG = 2.99.

Table 1. Ingredients and calculated composition of the dietary treatments for Experiment 1, as-fed basis.

	Diet					
Total Lysine, %	0.7	0.5	0.7	0.9	1.1	1.3
Ingredient, %	No RAC[a]					
Corn	85.06	91.40	85.06	78.67	72.26	65.81
Soybean meal-47.5% CP[b]	12.83	6.50	12.83	19.17	25.50	31.84
Biolys®[c]	0.18	0.13	0.18	0.23	0.28	0.33
L-threonine	0.03	0.00	0.03	0.08	0.12	0.17
DL-methionine	0.00	0.00	0.00	0.01	0.07	0.14
L-tryptophan	0.01	0.01	0.01	0.02	0.02	0.03
Dicalcium phosphate	0.52	0.56	0.52	0.48	0.44	0.39
Limestone	0.74	0.77	0.74	0.71	0.68	0.66
NaCl	0.25	0.25	0.25	0.25	0.25	0.25
Vitamin premix[d]	0.20	0.20	0.20	0.20	0.20	0.20
Trace mineral premix[e]	0.15	0.15	0.15	0.15	0.15	0.15
Antibiotic[f]	0.03	0.03	0.03	0.03	0.03	0.03
RAC, ppm	0.00	10.00	10.00	10.00	10.00	10.00
Calculated composition						
Lysine, %	0.70	0.50	0.70	0.90	1.10	1.30
CP, %	13.33	10.78	13.33	15.91	18.46	21.04
Calcium, %	0.45	0.45	0.45	0.45	0.45	0.45
Available phosphorus, %	0.15	0.15	0.15	0.15	0.15	0.15
ME[g], Mcal[h]/lb	1.52	1.52	1.52	1.52	1.52	1.52
True digestible AA[i]:Lys[j]						
Threonine	71.69	75.34	71.69	72.21	71.52	71.90
Met + Cys[k]	76.45	96.19	76.45	67.08	66.27	66.55
Tryptophan	20.92	22.01	20.92	21.57	20.97	21.41

[a]RAC = Ractopamine.
[b]CP = Crude protein.
[c]Biolys®, Evonik-Degussa Feed Additives (Kennesaw, Ga.).
[d]Supplied per kilogram of diet at 0.2% inclusion: vitamin A as retinyl acetate, 4,400 IU; cholecalciferol, 440 IU; α-tocopherol acetate, 24 IU; menadione sodium bisulfite, 3.5 mg; riboflavin 8.8 mg; d-pantothenic acid, 17.6 mg; niacin, 26.4 mg; vitamin B_{12}, 26.4 mg.
[e]Supplied per kilogram of diet at 0.15% inclusion: Zn (as $ZnSO_4$), 128 mg; Fe (as $FeSO_4 \bullet H_2O$), 128 mg; Mn (as MnO), 30 mg; Cu ($CuSO_4 \bullet 5\ H_2O$), 10.5 mg; I (as $Ca(IO_3) \bullet H_2O$), 0.26 mg; Se (as Na_2SeO_3), 0.26 mg.
[f]Tylan®, Elanco Animal Health (Greenfield, Ind.).
[g]ME = Metabolizable energy.
[h]Mcal = megacalories.
[i]AA = amino acid.
[j]Lys = lysine.
[k]Met + Cys = methionine plus cysteine.

Table 2. Repeated measure data for overall performance (4 weeks).

	Treatment							*P* values			
Lysine, %	0.7	0.5	0.7	0.9	1.1	1.3					
RAC[a], ppm	0	10	10	10	10	10	SEM[b]	TRT[c]	Wk[d]	T x W[e]	RAC
4 week											
ADG[f], lb	2.46	2.20	2.82	3.10	2.86	3.01	0.132	< 0.01	< 0.01	0.10	0.08
ADFI[g], lb	7.06	7.39	7.46	7.85	7.04	7.04	0.326	0.51	< 0.01	0.32	0.38
G:F[h], lb/lb	0.35	0.29	0.38	0.40	0.41	0.42	0.016	< 0.01	< 0.01	0.07	0.19
BFwg[i], in	0.06	0.07	0.07	0.10	0.06	0.06	0.012	0.23	< 0.01	0.47	0.49
LMAwg[j], in^2	0.36	0.26	0.34	0.47	0.40	0.41	0.070	0.45	< 0.01	0.25	0.84

[a]RAC = ractopamine.
[b]SEM = standard error of the mean.
[c]TRT = treatment.
[d]Wk = week.
[e]T x W = treatment by week interaction.
[f]ADG = average daily gain.
[g]ADFI = average daily feed intake.
[h]G:F = gain to feed ratio.
[i]BFwg = weekly gain in backfat thickness.
[j]LMAwg =weekly gain in longissimus muscle area.

Table 3. Broken line analysis for optimal total lysine concentration.

	Maximum response[a]	Requirement[b]	Upper 95%[c]	Sum of (residual^2)	R^2
4 week					
ADG[d]	2.99	0.76	0.89	0.535	51.5
G:F[e]	0.40	0.77	0.88	0.027	65.0

[a]Maximum response = the maximum response expected.
[b]Requirement = amount of total lysine, %, required in order to achieve the maximum response.
[c]Upper 95% = requirement plus 2 standard deviations of the mean requirement.
[d]ADG = average daily gain, lb.
[e]G:F = gain to feed ratio, lb/lb.

response was observed in pigs fed the diet containing the lowest amount of total lysine. There was a tendency for increased ($P < 0.08$) ADG of pigs fed 0.7% total lysine with ractopamine compared to pigs fed 0.7% total lysine without ractopamine. There were no significant differences detected for ADFI during the 4-week period ($P > 0.10$). Dietary treatment also had an effect on G:F ($P < 0.05$). Again, the lowest G:F was observed for pigs fed 0.5% total lysine. There was no significant effect of treatment on BF or LMA change during the experimental period ($P > 0.10$).

The four-week overall broken line regression data indicate that 0.76% total lysine was adequate in order for barrows to maximize ADG (Table 3). The results for G:F indicate a slightly greater requirement of 0.77% total lysine in order to maximize efficiency. It is important to note that requirements can be affected by several factors, including genetics and environmental conditions. The results of this experiment indicate that lysine concentration of the diet can affect the performance of pigs fed ractopamine. For barrows of the genetic background used, housed in experimental conditions, the broken-line regression results indicate that 0.76% total lysine is adequate for optimal growth performance.

Conclusions

Overall, dietary lysine concentration had significant effects on ADG and G:F. This indicates that total lysine concentration can play an important role in determining the amount of increased performance when feeding ractopamine. A total lysine concentration of 0.76% is adequate for obtaining maximal growth for barrows fed ractopamine. Further research is planned investigating the requirements of other amino acids when feeding ractopamine to finishing pigs.

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Effects of Lactose and Co-dried Milk-Yeast on Growth Performance and Gastrointestinal Health of Nursery Pigs

Lactose may affect the immune response mediated from the gut and interact with gastrointestinal microbiota; however no effects of milk-yeast on growth performance were detected.

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Summary

An experiment was conducted to evaluate the effects of dietary lactose alone or in combination with dried milk-yeast product on growth performance, gastrointestinal microbiota, and immune parameters in weanling pigs. Pigs fed lactose and lactose with milk-yeast tended (P = 0.07) to have greater BW compared to control pigs (19.56 and 19.60 vs. 18.55 lb) at the end of phase 1 (week 1 to 2); however, no differences in BW were observed during phase 2 (week 3 to 4), phase 3 (week 5), or overall (week 1 to 5). With respect to growth performance, pigs fed lactose and lactose plus milk-yeast had greater (P = 0.05) ADG, and tended (P = 0.07) to have greater ADFI compared to control pigs during phase 1. There were no differences observed for ADG or ADFI during phase 2, 3, or the overall experimental period. With respect to immune parameters, a main effect of treatment was observed for circulating immunoglobulin (Ig)A where control pigs had greater (P < 0.01) concentrations of IgA compared to pigs fed lactose with or without milk-yeast; however, no effects of dietary treatment were observed for circulating IgG or tumor necrosis factor alpha. Lastly, fecal microbiota of control pigs had a greater microbial diversity index (Shannon's, P = 0.03) compared to pigs fed lactose plus milk-yeast on day 0; however, no differences in microbial diversity indices were observed on days 7 or 14 among dietary treatments. In addition, a shift in microbial composition, limited to a small number of microbial groups, was observed on day 7 with lactose fed pigs having greater (P = 0.05) putative L. jonhsonii *staining intensity compared to control pigs and pigs fed lactose plus milk-yeast. On day 14,* L. reuteri *tended (P = 0.15) to be enhanced, and* L. delbrueckii *was virtually eliminated (P = 0.04) by feeding lactose with or without milk-yeast. This research indicates that growth performance, immune parameters, and composition of the fecal microbiota may be affected by dietary inclusion of lactose alone or in combination with milk-yeast.*

Introduction

Stressors at weaning (including dietary, environmental, and social stressors) lead to the reduction of feed intake, nutrient absorption (due to villous atrophy in conjunction with a greater incidence of diarrhea), and consequently, decrease the overall growth performance of weaned pigs. Feeding lactose has been shown to increase feed intake and feed efficiency in weanling pigs. Although inconsistent results exist, there is evidence that dietary lactose may be a potential prebiotic for nursery pigs and may have a positive effect on modulating gut microbiota. In the gastrointestinal tract (GIT), lactose is fermented to lactic acid by lactic acid-producing bacteria (resulting in a decrease in pH) which is not favorable for pathogenic bacteria but may promote the proliferation of commensal bacteria. Furthermore, the inclusion of lactose may increase the production of short-chain fatty acids, particularly butyric acid, which are important sources of energy for gut epithelial cells. In addition, supplementation of yeast culture or live yeast in nursery diets has resulted in positive effects on growth performance and gut health of pigs. Previous research showed that pigs fed yeast had increased beneficial bacteria (i.e., lactobacilli) and reduced coliform bacteria in the small intestine. Yeast mixtures contain a variety of active components such as enzymes, hormones, nucleic acids, and cell wall products (e.g., mannanoligosaccharides and beta glucans) that may be beneficial to the host.

There has been considerable focus on research evaluating the effects of lactose and yeast individually; however, the effect of feeding lactose in combination with milk-yeast on the growth performance and gastrointestinal health of nursery pigs has not been investigated. Therefore, the objective of the current study was to determine effect of lactose and lactose in combination with milk-yeast on growth performance, gastrointestinal microbiota, and immune parameters of nursery pigs.

Table 1. Composition of experimental diets (as-fed basis).

	Phase 1			Phase 2			Phase 3		
Treatments[1]	A	B	C	A	B	C	A	B	C
Lactose, %	0	20	20	0	15	15	0	5	5
Milk-yeast, %	0	0	5	0	0	5	0	0	5
Ingredient, %									
Corn	61.500	37.085	33.408	61.460	43.140	39.510	58.760	52.640	48.968
Soybean meal, 46.5% CP	20.5	20.5	20.5	22	22	22	28.75	28.75	28.75
Spray-dried porcine plasma	5	5	5	2.5	2.5	2.5	0	0	0
Select menhaden fish meal	6	6	6	7.5	7.5	7.5	6	6	6
DairyLac 80	0	25	23.913	0	18.750	17.600	0	6.250	5.163
Dicalcium phosphate, 18.5% P	1.325	0.500	0.295	0.750	0.150	0.000	0.900	0.700	0.500
Limestone	0.250	0.400	0.475	0.300	0.390	0.430	0.300	0.350	0.420
Salt	0.3	0.3	0.3	0.3	0.3	0.3	0.4	0.4	0.4
Zinc oxide	0.3	0.3	0.3	0.3	0.3	0.3	0	0	0
Vitamin premix[2]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Trace mineral premix[3]	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
L-Lysine•HCl	0.210	0.210	0.080	0.265	0.275	0.145	0.255	0.260	0.130
DL-Methionine	0.120	0.180	0.195	0.115	0.160	0.170	0.125	0.130	0.145
L-Threonine	0.095	0.125	0.135	0.110	0.135	0.145	0.110	0.120	0.125
Mecadox 2.5	1	1	1	1	1	1	1	1	1
Corn oil	3	3	3	3	3	3	3	3	3
Milk-yeast	0	0	5	0	0	5	0	0	5

[1]Dietary treatments included: control (A; no Dairylac 80 and milk-yeast); Dairylac 80 (B); and Dairylac 80 supplemented with 5% milk-yeast (C).
[2]Vitamin premix containing: vitamin A as retinyl acetate, 5,500 IU; vitamin D_3 as cholecalciferol, 550 IU; vitamin E as alpha-tocopherol acetate, 30 IU; vitamin K as menadione dimethylpyrimidinol bisulfide, 4.4 mg/kg; niacin, 33 mg/kg, pantothenic acid as d-Calcium pantothenate, 22.05 mg/kg; riboflavin, 11 mg/kg; vitamin B12 as cyanocobalamin, 33 mg/kg.
[3] Trace mineral premix containing: copper (as $CuSO_4 \bullet 5H_2O$), 10 mg/kg; iodine (as Ca $(IO_3) \bullet H_2O$), 0.25 mg/kg; Iron ($FeSO_4 \bullet 2H_2O$), 125 mg/kg; manganese (MnO), 15 mg/kg; Selenium (Na_2SeO_3), 0.3 mg/kg; Zinc ($ZnSO_4 \bullet H_2O$), 125 mg/kg.

Materials and Methods

Animals and Experimental Design

The experimental protocol was reviewed and approved by the Institutional Animal Care and Use Committee of the University of Nebraska–Lincoln. One hundred eight weaned pigs (20 ± 1 day of age) were sorted by initial body weight (BW) and sex, and randomly allotted to dietary treatment (three treatments; six pigs/pen). The average initial body weight was 13.33 ± 0.07 lb and there were six replicates/treatment with three barrows and three gilts per pen. Pigs were housed in a temperature-controlled room and each pen had a single nipple waterer and a single self-feeder for ad libitum access to water and feed. The study consisted of a five-week feeding experiment divided into three phases: phase 1 (week 1 to 2), phase 2 (week 3 to 4), and phase 3 (week 5).

Dietary Treatments

The ingredient composition and calculated analysis of experimental diets are presented in Tables 1 and 2, respectively. Dietary treatments included the following: A) Control (CTL; no Dairylac 80 or milk-yeast); B) Dairylac 80; and C) Dairylac 80 supplemented with milk-yeast (5%). Phase 1, 2 and 3 diets were formulated to contain 1.47, 1.42, and 1.37 true ileal digestible Lys, respectively. Total Lys was 1.56% in phase 1 and 2, and 1.51% in Phase 3 diets. Except for the control diet, dietary treatments in phase 1, 2, and 3 contained a total of 20, 15 and 5% lactose, respectively. Diets were formulated to meet or exceed NRC (1998) requirements. Dairylac 80 and co-dried milk-yeast 5050 (International Ingredient Corp., St. Louis, Mo.) were the sources of lactose and milk-yeast used in experimental diets. Dairylac 80 produced from sweet and dried whey soluble was a granular and nonhygroscopic product, and contained 3.2% CP, and 0.06% Lys (analyzed composition) and 80% lactose. Co-dried milk-yeast was produced from 50% dried near-dated-milk and 50% dried brewer's yeast containing 17.4% lactose, 33% CP and 1.82% Lys (analyzed composition). This milk-yeast product was included in treatment C during all three feeding phases.

Data and Sample Collection

Individual pig weights and feed disappearance were recorded on days 0, 7, 14, 21, 28, and 35 to calculate average daily gain (ADG), average daily feed intake (ADFI), and feed efficiency (ADG:ADFI). Blood samples were collected from all pigs via jugular venipuncture. Serum was harvested following centrifugation (20 min at

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1,500 × *g*). Two pigs (one gilt and one barrow) in each pen were randomly selected for collection of fecal samples. Blood and fecal samples were collected at timepoints that coincided with pig BW measurements. Serum and fecal samples were frozen at -20°C for subsequent analyses.

Laboratory Analysis

A porcine specific, enzyme-linked immunosorbent assay (ELISA) was used to quantify circulating immunoglobulin (Ig) G and A (Bethyl Laboratories, Inc., Montgomery, Tex.), and tumor necrosis factor (TNF)-α (R&D Systems, Minneapolis, Minn.).

Isolation of fecal DNA was conducted as described by Martinez et al (2009). The resultant DNA was utilized for subsequent polymerase chain reaction (PCR) and denaturing gradient gel electrophoresis (DGGE) analyses. Briefly, for investigation of the entire microbe population in fecal samples, PCR was performed by using universal primers to amplify the V3 region of 16S rRNA gene. In addition, to analyze specific *Lactobacillus* populations in fecal samples, PCR was performed by using the lactic acid bacteria specific primers. Images obtained from DGGE were evaluated using the BioNumerics software and staining intensities of individual bands were determined as proportion of peak surface area relative to the surface area of the entire molecular fingerprint of the sample. Diversity of microbiota was calculated by Shannon's and Simpson's indices using the following formulas:

$$\text{Shannon's index} = \sum_{i=1}^{n} -pi \ln(pi)$$

$$\text{Simpson's index} = \sum_{i=1}^{n} \frac{-ni(ni-1)}{N(N-1)}$$

In which, n*i* was the number of organisms belonging to species *i* (as proportion of band intensity in respect to entire intensity of fingerprint); N was the total number of organisms in microbial population; p*i* was the proportion of a species i present in a sample. The larger Shannon's index, the more diverse the microbial population. Conversely, the smaller the Simpson's index, the more diverse the microbial population.

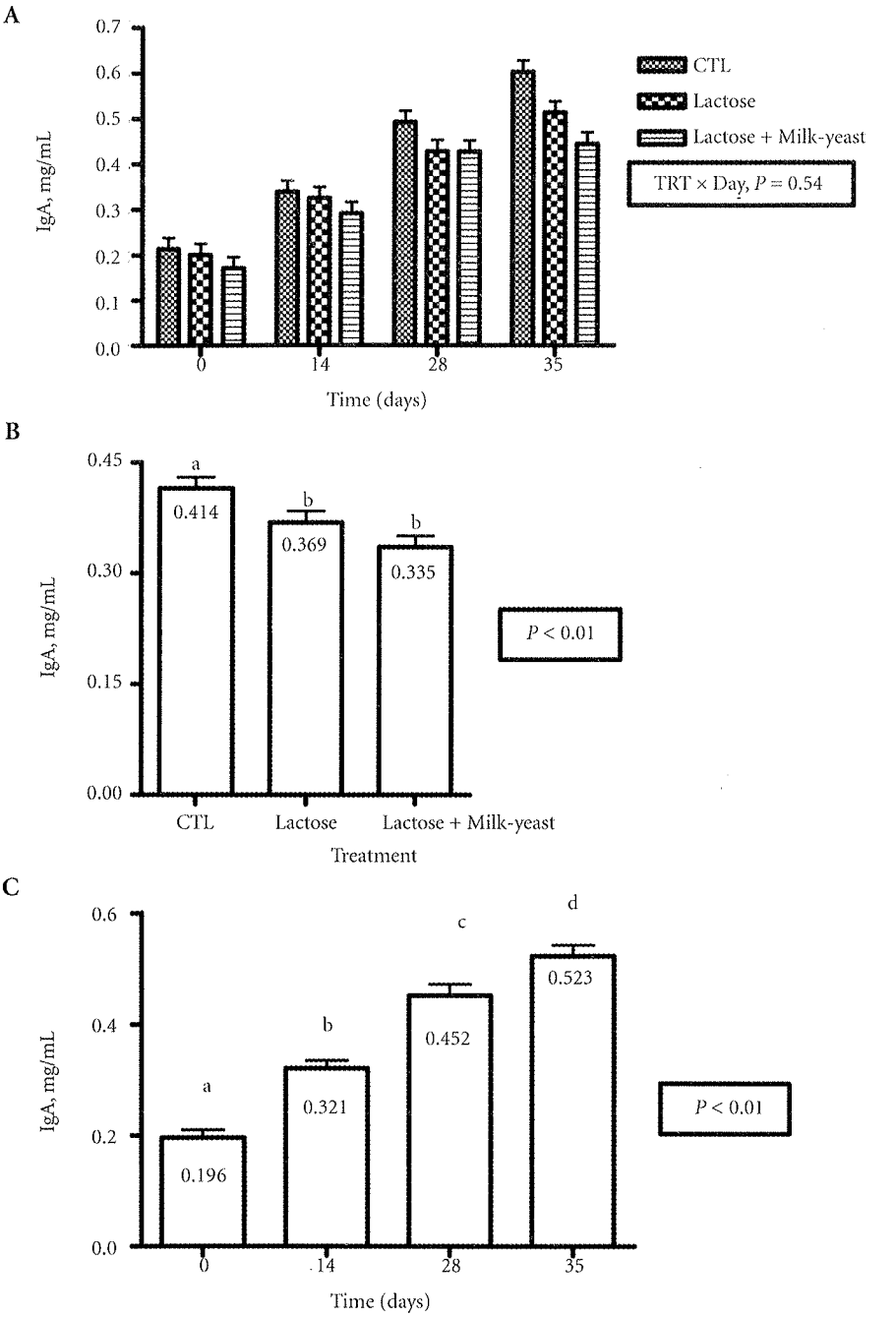

Figure 1. Effects of feeding lactose and milk-yeast on circulating concentrations of Immunoglobulin (Ig) A in weanling pigs. Each bar represents the least-squares mean (± SEM) of 36 (for days 0 and 14) and 18 (for days 28 and 35) observations. Bars with different superscripts differ at $P < 0.05$; panel A represents the interactive treatment × day means, panel B represents treatment means, and panel C represents day means.

Statistical Analysis

Each pen was considered as an experimental unit. Data were analyzed as a completely randomized design using the MIXED procedure of SAS. Pen was considered a random effect. All means are presented as least-squares means.

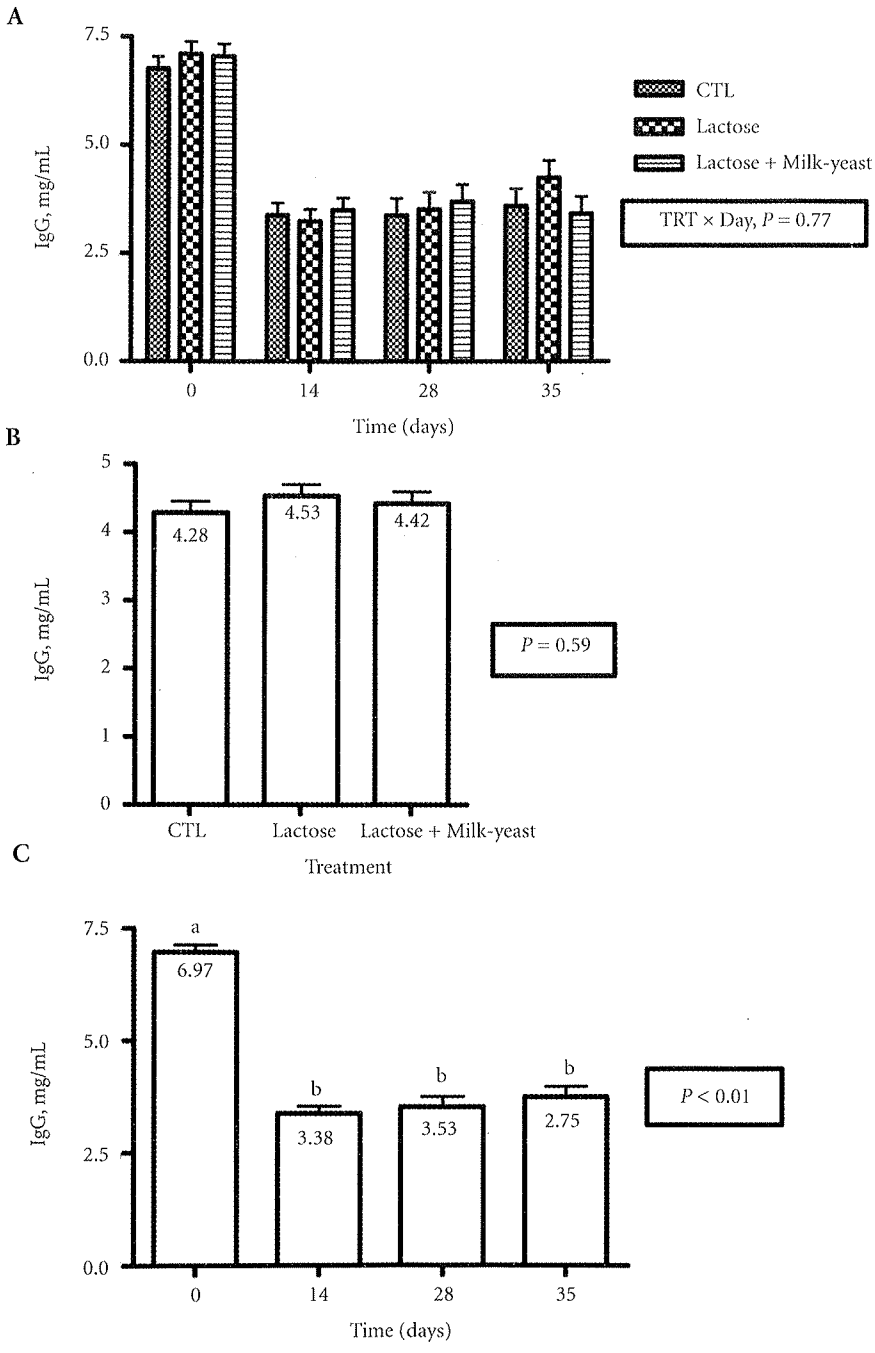

Figure 2. Effects of feeding lactose and milk-yeast on circulating concentrations of Immunoglobulin (Ig) G in weanling pigs. Each bar represents the least-squares mean (± SEM) of 36 (for days 0 and 14) and 18 (for days 28 and 35) observations. Panel A represents interactive treatment × day means, panel B represents treatment means, and panel C represents day means.

Results and Discussion

Growth Performance

Pigs fed lactose with or without milk-yeast tended to have greater BW at the end of week 1 ($P = 0.09$) and 2 ($P = 0.07$) compared to the CTL pigs. There were no effects of dietary treatment on pig BW during week 3, 4, 5, or overall (week 1 to 5).

Compared to CTL pigs, pigs fed lactose and milk-yeast had greater ($P = 0.01$) ADG and ADG:ADFI during week 1 and increased ($P = 0.05$) ADFI during week 2. During phase 1 (week 1 to 2), pigs fed lactose supplemented with milk-yeast had greater ($P = 0.05$) ADG and tended ($P = 0.07$) to have greater ADFI compared to CTL pigs. No differences in ADG, ADFI, or ADG:ADFI were observed between pigs fed lactose supplemented with and without milk-yeast during phase 1 (week 1 to 2).

During phase 2, there were no differences among treatments for ADG, ADFI and ADG:ADFI; however, CTL pigs tended ($P = 0.08$) to have greater ADG:ADFI compared to lactose fed pigs with or without milk-yeast during phase 2 (week 3 to 4). There were no effects of treatment on growth performance during phase 3 (week 5). Overall (week 1 to 5), there were no effects of dietary treatment on growth performance. These results agree with previous research that reported the inclusion of lactose had positive effects on growth performance during the early nursery phases. In summary, the traditional effects of lactose on phase-1 nursery performance were observed. However, there were no effects of milk-yeast product on growth performance.

Immune parameters: IgA, IgG, and tumor necrosis factor (TNF) α

There were no observed treatment × time (day) interactions observed for any of the immune parameters evaluated (Figures 1A, 2A, and 3A). However, significant main effects of treatment (IgA; Figure 1B) and time (IgA, IgG, and TNF-α; Figures 1C, 2B, and 3B, respectively) were observed. Specifically, when means were averaged among all timepoints, CTL pigs had greater ($P < 0.01$) circulating IgA compared to pigs fed lactose with or without milk-yeast. Dietary lactose is considered as a prebiotic for pigs and may improve gut barrier function by facilitating mucus secretion and tight junction formation, by providing immunologic factors, and by lowering GIT pH which is unfavorable for pathogenic bacteria. Thus, there is a

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possibility that a decreased pathogenic load on the GIT exists in pigs fed diets containing lactose with or without milk-yeast compared to the pigs fed no lactose or milk-yeast. In addition, circulating IgA increased ($P < 0.01$) with time throughout the duration of the experiment. This result may be explained by the maturation of the piglet immune system. Previous research indicates that maternal IgA predominates in piglet serum until approximately 5 weeks of age, at which time the first IgA-bearing B cells are evident in the lamina propria of the GIT wall and IgA becomes the predominant Ig isotype in the GIT.

With respect to IgG, pigs were raised in a clean research environment with no clinical signs of disease which may explain the lack of any effects among dietary treatments. However, a main effect of time ($P < 0.01$) was observed with IgG present at the greatest concentration at weaning (day 0) and lower concentrations present throughout the duration of the experiment. Again, as with IgA, this may be explained by the residual concentrations of IgG present in the piglets as a result of passive transfer from the dam and the inability for young pigs to synthesize their own IgG until they are more mature.

Tumor necrosis factor-α is a proinflammatory cytokine which is synthesized and secreted during stress, endotoxemia, or disease. The concentration of TNF-α in piglets in the current experiment was only measured on days 0 and 14. There were no main effects of treatment on TNF-α; however, when means were averaged among treatments, pigs had greater ($P < 0.05$) concentrations of TNF-α at weaning (day 0) compared to day 14. It is speculated that stress due to weaning may be responsible for the increase in this proinflammatory cytokine at day 0.

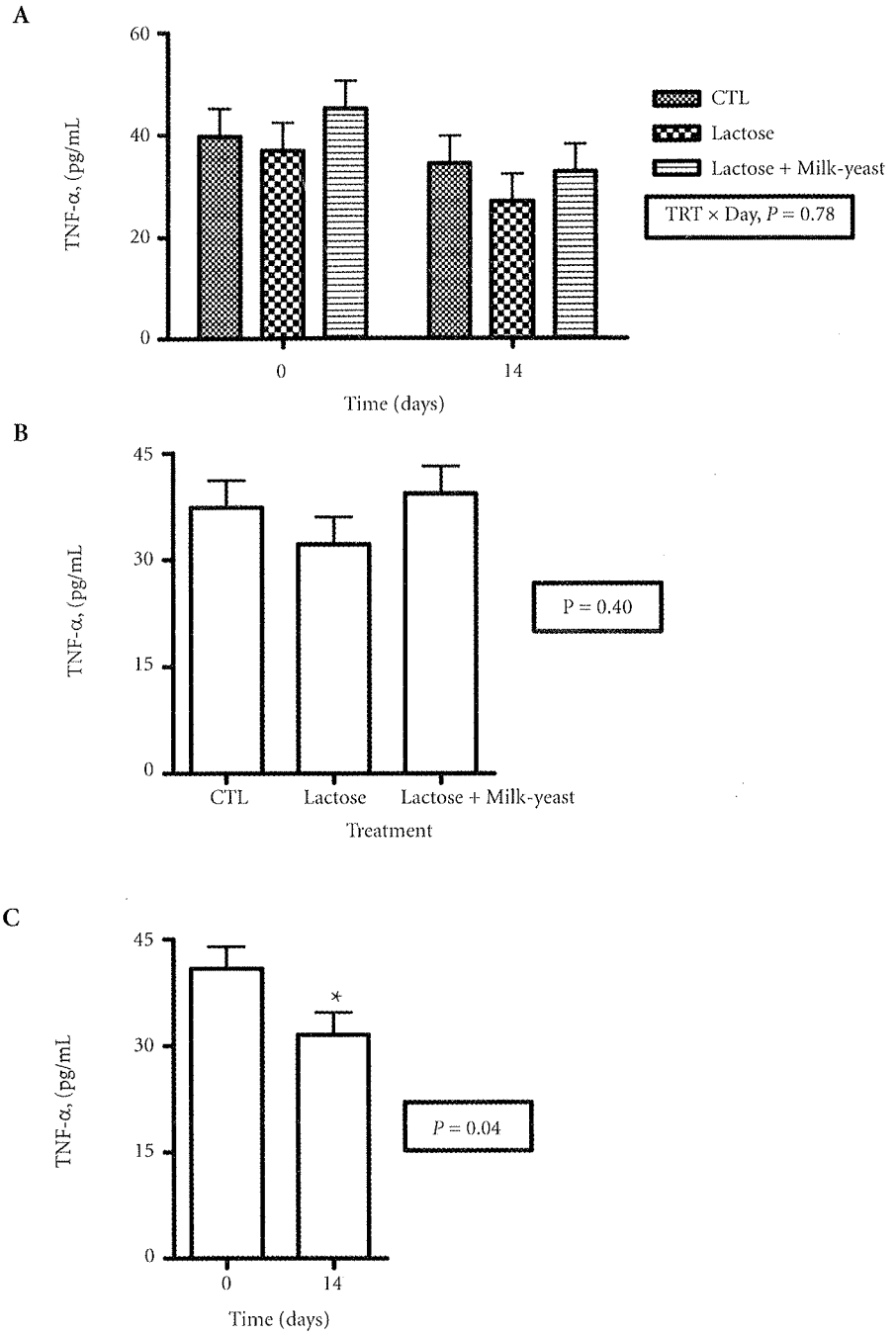

Figure 3. Effects of feeding lactose and milk-yeast on circulating concentration of tumor necrosis factor (TNF)-α. Each bar represents the least-squares mean (+ SEM) of 24 observations. Panel A represents interactive treatment × day means; Panel B represents treatment effects; And panel C represents day effects.

DGGE Analysis of the Lactobacillus Biota in Fecal Samples

The ratio of staining intensity of dominant bands as a proportion of total fingerprint intensity using *Lactobacillus*-targeted primers is summarized in Table 4. At weaning (day 0), there were no differences among treatments on fecal *Lactobacillus* staining intensity. Bands corresponding to *L. johnsonii* (range: 17.68 to 20.68%), *L. sobrius/amylovorus* (range: 6.15 to 11.39%), *and L. mucosae* (range: 11.74 to 17.93%) in the marker are the most dominant Lactobacilli present in pigs at weaning. Bands corresponding to *L. reuteri* appeared at a lower proportion at weaning (5.16 to 9.7%).

At day 7 postweaning, pigs fed

Table 2. Calculated and determined diet composition (as-fed basis).

	Phase 1			Phase 2			Phase 3		
Treatment[1]	A	B	C	A	B	C	A	B	C
Lactose, %	0	20	20	0	15	15	0	5	5
Milk-yeast, %	0	0	5	0	0	5	0	0	5
Calculated analysis									
CP,[2] %	22.8	22	23.1	22.5	22	23.1	22.5	22.3	23.5
Total lys, %	1.57	1.56	1.56	1.56	1.56	1.56	1.51	1.51	1.51
Tid[3] lys, %	1.47	1.47	1.47	1.42	1.42	1.42	1.37	1.37	1.37
Ca, %	0.85	0.85	0.85	0.82	0.82	0.82	0.8	0.8	0.8
P, %	0.81	0.75	0.74	0.73	0.68	0.69	0.71	0.7	0.69
Available P, %	0.53	0.53	0.53	0.44	0.44	0.45	0.4	0.4	0.4
ME[4], kcal[5]/lb	1,532	1,529	1,543	1,534	1,532	1,547	1,529	1,528	1,543
Determined analysis									
CP, %	21.68	21.05	22.14	21.75	20.78	22.36	22.10	21.70	22.86
Ether extract, %	6.30	5.36	5.71	6.32	5.79	6.13	6.32	6.02	6.32
Lys, %	1.43	1.46	1.37	1.42	1.39	1.44	1.40	1.42	1.41
Met, %	0.46	0.52	0.49	0.48	0.50	0.52	0.48	0.48	0.52
Thr, %	0.92	0.94	1.0	0.94	0.94	1.01	0.89	0.89	0.94

[1]Dietary treatments included: control (A, no Dairylac 80 and milk-yeast); Dairylac 80 (B); Dairylac 80 supplemented with 5% dried milk-yeast (C).
[2]Crude protein.
[3]True ileal digestible.
[4]Metabolizable energy.

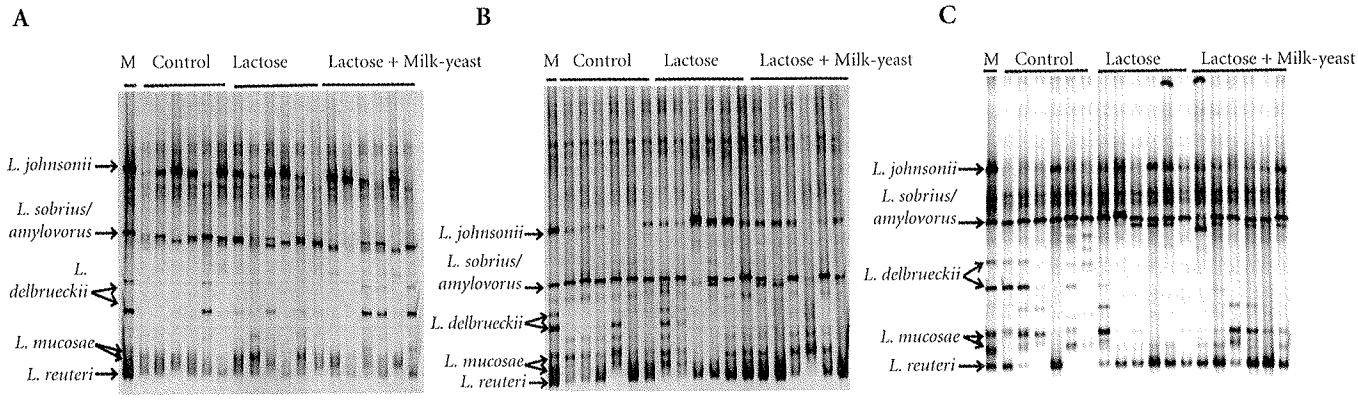

Figure 4. Effects of feeding lactose and milk-yeast on *Lactobacillus* diversity determined by DGGE using *Lactobacillus*-targeted primers. Each lane contains DNA isolated from fecal samples obtained from individual pigs (n = 18); Lane *M is* a marker containing reference *Lactobacillus* species commonly present in pigs. Panel A represents fecal samples of pigs at weaning. Panel B represents fecal samples of pigs at 7 days after weaning. Panel C represent fecal samples of pigs at 14 days after weaning.

lactose with (18.17%) or without milk-yeast (7.63%) had greater (P = 0.05) staining intensity of the putative *L. johnsonii* when compared to the CTL pigs (4.64%). Previous data suggest that 16 to 89% of *L. johnsonii* can utilize lactose and this may explain the increase of this species in pigs fed lactose for 7 days. Interestingly, *L. johnsonii* is believed to be a probiotic species with ability to produce hydrogen peroxide *in vitro* and contribute to the defense against pathogen infection. There were no differences among other *Lactobacillus* species at day 7; however, the tendency of one unknown species (P = 0.08) did appear in pigs fed lactose with or without yeast, but were absent in CTL pigs. There were changes in *Lactobacillus* composition at day 7 where *L. sobrius/amylovorus* (range: 14.11 to 26.33%) and *L. reuteri*

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(range: 17.52 to 20.65%) became the most dominant species compared to day 0 (range: 6 to 10% and 5 to 9%, respectively for *L. sobrius/amylovorus* and *L. reuteri*). The increasing abundance of these two species may be a reason for the reduction of average staining intensity of other species such as *L. johnsonii* at day 7 (10.15%) compared to day 0 (19.51%). In addition, *L. johnsonii* is known as a casein-utilizing species; therefore, the change of protein supply from sow's milk to protein in the nursery diet may result in decreasing casein available and reduction of species utilizing this protein at day 7.

At day 14 postweaning, the CTL pigs had greater putative *L. sobrius/amylovorus* ($P = 0.10$) and *L. delbrueckii* ($P = 0.04$). Otherwise, there were no differences among treatments on other *Lactobacillus* species. However, putative *L. reuteri* was numerically greater in pigs fed lactose with yeast (16.56%) or without milk-yeast (12.5%) compared to the control pigs (7.82%). This is a significant finding because *L. reuteri* has been reported to confer health benefits to humans and animals and is commonly used as a probiotic bacteria for pigs. In summary, feeding lactose with or without milk-yeast did affect the maintenance of some *Lactobacillus* species which are probiotic species (*L. reuteri* and *L. johnsonii*) in pigs after weaning.

DGGE Analysis of the Total Fecal Microbiota by DGGE

The effects of feeding lactose and milk-yeast on total microbial composition were determined by the staining intensity and diversity indexes (Shannon's and Simpson's) using PCR-DGGE in combination with universal primers (Table 6). At days 0, 7, and 14, there were no differences among treatments on microbial population based on Shannon's and Simpson's diversity indices. However, a greater ($P = 0.03$) Shannon's index was identified for control pigs at weaning, compared lactose-fed pigs with and

Table 3. Effects of feeding lactose and milk-yeast on pig performance.

	Treatment[1]				
Item	A	B	C	SEM[2]	*P*-value
BW, lb					
Week 0	13.38	13.38	13.31	0.07	0.75
Week 1	14.15	14.45	14.63	0.15	**0.09**
Week 2	18.55	19.56	19.60	0.33	**0.07**
Week 3	26.11	27.63	27.10	0.53	0.15
Week 4	35.31	36.98	36.37	0.66	0.22
Week 5	46.35	48.05	47.85	0.84	0.33
Phase 1 (week 1)					
ADG, lb	0.11[a]	0.16[b]	0.19[b]	0.02	**0.01**
ADFI, lb	0.25	0.26	0.28	0.01	0.2
G:F, lb/lb	0.44[a]	0.61[b]	0.68[b]	0.05	**0.01**
Phase 1 (week 2)					
ADG, lb	0.63	0.70	0.71	0.04	0.24
ADFI, lb	0.75[a]	0.86[b]	0.87[b]	0.03	**0.05**
G:F, lb/lb	0.84	0.82	0.82	0.02	0.87
Phase 1 (week 1,2)					
ADG, lb	0.37[a]	0.43[b]	0.45[b]	0.02	**0.05**
ADFI, lb	0.50	0.56	0.57	0.02	**0.07**
G:F, lb/lb	0.74	0.77	0.79	0.02	0.34
Phase 2 (week 3)					
ADG, lb	1.08	1.15	1.07	0.04	0.35
ADFI, lb	1.44	1.60	1.52	0.05	0.14
G:F, lb/lb	0.76	0.72	0.70	0.01	**0.07**
Phase 2 (week 4)					
ADG, lb	1.31	1.34	1.32	0.03	0.87
ADFI, lb	1.93	2.04	1.96	0.05	0.3
G:F, lb/lb	0.68	0.65	0.67	0.01	0.14
Phase 2 (week 3,4)					
ADG, lb	1.20	1.25	1.20	0.03	0.48
ADFI, lb	1.68	1.82	1.74	0.05	0.18
G:F, lb/lb	0.71	0.68	0.69	0.01	**0.08**
Phase 3 (week 5)					
ADG, lb	1.58	1.58	1.64	0.05	0.57
ADFI, lb	2.30	2.36	2.38	0.05	0.49
G:F, lb/lb	0.69	0.67	0.69	0.02	0.63
Overall (week 1 to 5)					
ADG, lb	0.94	0.98	0.99	0.02	0.42
ADFI, lb	1.33	1.41	1.40	0.04	0.25
G:F, lb/lb	0.71	0.69	0.70	0.01	0.51

[1]Dietary treatments included: control (A; no Dairylac 80 and milk-yeast); Dairylac 80 (B); and Dairylac 80 supplemented with 5% milk-yeast (C).
[2]Standard error of the mean.
[ab]Means in the same row with different superscript differ ($P < 0.05$).

Table 4. Ratio of staining intensities[1] of dominant bands as a proportion of total fingerprint intensity using *Lactobacillus* primer specific DGGE.

Day	Band	Putative species	Control	Lactose	Lactose + Milk-yeast	SEM	*P*
0	1	*L. johnsonii*	20.2	17.7	20.7	6.6	0.94
	2	*L. sobrius/amylovorus*	10.6	11.4	6.15	3.4	0.52
	3	*L. delbrueckii*	2.08	1.50	8.91	2.7	0.12
	4	*L. mucosae*	15.6	17.9	11.7	3.9	0.55
	5	*L. reuteri*	9.7	5.16	5.95	2.4	0.37
7	1	*L. johnsonii*	4.64[a]	18.17[b]	7.63[a]	3.8	**0.05**
	2	*L. sobrius/amylovorus*	26.3	16.2	14.1	4.2	0.12
	3	unknown	0.00[a]	3.39[b]	2.20[ab]	1.0	**0.08**
	4	*L. delbrueckii*	1.82	2.47	2.07	1.8	0.97
	5	*L. mucosae*	14.3	7.00	15.2	4.6	0.40
	6	*L. reuteri*	20.7	18.7	17.5	5.4	0.92
14	1	*L. johnsonii*	14.0	17.2	12.3	2.5	0.38
	2	*L. sobrius/amylovorus*	20.5[a]	17.9[ab]	14.4[b]	1.8	**0.10**
	3	*L. delbrueckii*	7.77[a]	0.10[b]	0.00[b]	2.2	**0.04**
	4	*L. mucosae*	8.33	3.87	9.32	2.7	0.33
	5	*L. reuteri*	7.82	12.5	16.6	3.6	0.26
	6	unknown	2.70	5.90	5.51	1.9	0.44

[1]Intensity of individual bands as determined as percentage of the peak surface area relative to the surface area of the entire molecular fingerprint of the sample; pigs were at weaning (day 0), 7 and 14 days after weaning. Each value represents a least squares mean of 6 pigs per treatments.
[ab]Means in the same row with different superscript differ ($P < 0.1$)

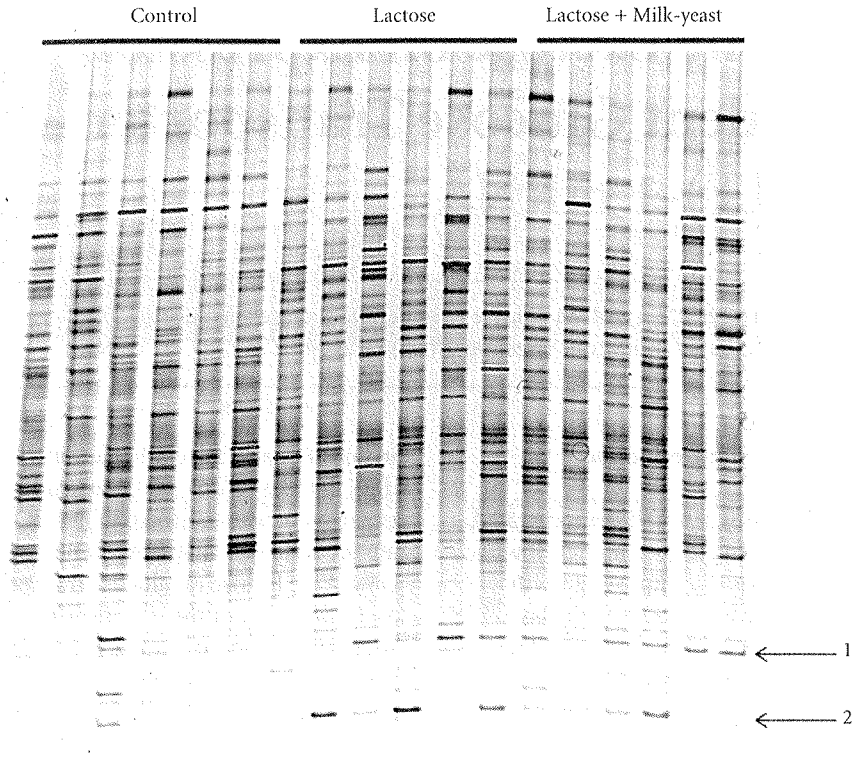

Figure 5. Effects of feeding lactose and milk-yeast on diversity of total microbiota population determined by DGGE using universal PCR primers. Each lane contains DNA isolated from fecal samples obtained from individual pigs (n = 18) at 7 days post weaning. The arrows indicated the bands whose staining intensity was increased (Table 5).

Table 5. Ratio of staining intensities[1] of dominant bands as a proportion of total fingerprint intensity using universal primer DGGE.

Day	Band	Control	Lactose	Lactose + Milk-yeast	SEM[1]	*P*
7	1	0.27[a]	1.31[b]	1.58[b]	0.33	**0.04**
	2	0.31	1.81	0.88	0.56	0.19

[1]Standard error of the mean.
[ab]Means in the same row with different superscript differ ($P < 0.05$).

Table 6. Effects of feeding lactose and milk-yeast on diversity of fecal microbiota.

Day	Treatment	Shannon's[1] index (mean ± SEM[3])	Simpson's[2] index (mean ± SEM)	*P*-value Shannon	*P*-value Simpson
	Control	3.44 ± 0.04[a]	0.045 ± 0.003		
0	Lactose	3.33 ± 0.04[ab]	0.051 ± 0.003	0.03	0.14
	Lactose + Milk-yeast	3.24 ± 0.04[b]	0.055 ± 0.003		
	Control	3.49 ± 0.58	0.039 ± 0.002		
7	Lactose	3.50 ± 0.58	0.404 ± 0.002	0.97	0.88
	Lactose + Milk-yeast	3.57 ± 0.58	0.039 ± 0.002		
	Control	2.93 ± 0.096	0.084 ± 0.013		
14	Lactose	2.91 ± 0.096	0.091 ± 0.013	0.69	0.52
	Lactose + Milk-yeast	3.02 ± 0.096	0.069 ± 0.013		

[1]The larger the Shannon's index, the more diverse the microbial population.
[2]The smaller the Simpson's index, the more diverse the microbial population.
[3]Standard error of the mean.
[ab]Means in the same column with different superscript differ ($P < 0.05$). Each value represents a least squares mean of 6 observations.

without milk-yeast. In addition, DGGE analysis revealed changes in the composition of the fecal microbiota in as much as two bands were significantly more common in animals fed lactose and lactose with yeast at day 7 (Figure 5, bands 1 and 2). Pigs fed lactose with or without yeast had greater ($P = 0.04$) staining intensities of a bacteria type aligned at band 1 in comparison with CTL pigs (Table 5). This dietary effect may relate to the change of *Lactobacillus* composition in pigs at the same age as presented in Table 6.

Conclusions

During phase 1, pigs fed lactose with and without milk-yeast had increased growth performance compared to CTL pigs; however, this effect of lactose and milk-yeast did not continue into the latter phases of the nursery stage. In addition, lactose and milk-yeast may have positive effects on the health status of weanling pigs; however, more research is needed to examine the interplay among growth performance, health, and microbial populations of young pigs fed prebiotics such as lactose and milk-yeast.

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Effects of Crystalline Lactose on Expression of Sodium-dependent Glucose Transporter (SGLT)-1 mRNA

Supplementation of crystalline lactose increases SGLT-1 mRNA relative abundance, glucose transport to the basolateral compartment, and reduces the LPS-mediated inhibitory effect on glucose transport in IPEC-J2 cells.

Huyen Tran
Phillip S. Miller
Thomas E. Burkey[1]

Summary

A preliminary experiment was conducted to evaluate the effects of crystalline lactose on the relative abundance of sodium-dependent glucose transporter (SGLT)-1 mRNA in vitro in model porcine jejunal epithelial cells (IPEC-J2). Cells were treated with low (28 mM) or high (56 mM) concentrations of lactose alone or in combination with lipopolysaccharide (LPS; 10 ng/mL). Total RNA and culture media samples from both apical and basolateral compartments were harvested at 3, 6, 12, and 24 hours following the addition of respective treatments. With respect to relative abundance of SGLT-1 mRNA, there were no interactions of lactose, LPS, and time; however, a main effect of lactose ($P < 0.01$) was observed. Cells treated with a high concentration of lactose had greater SGLT-1 mRNA expression compared to the control cells ($P = 0.003$) and low concentration lactose ($P = 0.02$) treated cells. With respect to glucose quantification, polarization of glucose in the basolateral rather than in the apical compartment ($P = 0.04$) was observed in a time-dependent manner ($P < 0.001$). Supplementation of lactose in LPS-treated cells reduced LPS-mediated inhibitory effect on glucose transport from the apical to basolateral compartment ($P = 0.07$).

Introduction

Previous research demonstrated that including lactose in diets for nursery pigs can improve growth performance and gut health (i.e., stimulating epithelial cell proliferation and mediating beneficial bacteria colonization in the gastrointestinal tract (GIT). However, molecular mechanisms for how lactose affects pig growth remain unclear. In the small intestine, lactose is hydrolyzed and absorbed as glucose and galactose via the action of lactase present along the brush border membrane. The absorption of glucose and galactose involves two steps mediated by the sodium-dependent glucose transporter (SGLT)-1 and glucose transporter (GLUT)-2. First, sugar transport is coupled to sodium and electrical gradients across the epithelial cell membrane providing energy for the influx of the sugars. This step is facilitated by SGLT-1 (an integral protein in the brush border membrane). Second, glucose and galactose are transported across the basolateral membrane of enterocytes to the blood via GLUT-2. The dissociation of Na^+ at the cytosolic surface is the rate-limiting step of SGLT-1 transport. Thus, SGLT-1 plays an important role in sugar uptake from the GIT lumen to enterocytes. Research studies have investigated effects of different sugars, fiber, and sodium on SGLT-1 gene expression; however, a mechanism for how lactose affects expression of SGLT-1 mRNA in cultured porcine epithelial cells does not exist. Thus, a preliminary experiment was conducted to evaluate the effects of supplementation of crystalline lactose on glucose transport and expression of mRNA encoding for sodium-dependent glucose transporter (SGLT)-1 *in vitro* in model porcine jejunal epithelial cells (IPEC-J2).

Materials and Methods

IPEC-J2 Cell Cultures and Treatment Design

Porcine jejunal intestinal epithelial cells have been characterized previously and are non-transformed, jejunal epithelial cells derived from neonatal pigs, and are maintained as a continuous culture. Cell cultures were maintained in DMEM-F12 growth medium supplemented with insulin/transferin/Na selenite media supplement, epidermal growth factor, antibiotic, and fetal bovine serum. For experimentation, IPEC-J2 cells were seeded onto 12-well transwell cell culture plates and maintained in the above media. The cells were incubated for 24 hours before being washed and re-fed every other day for seven days to form a model of the gut epithelium. Twenty-four hours before experimentation, cells were washed and re-fed media devoid of antibiotics.

There were six treatments (2 × 3 factorial) included in this experiment: 1) control (CTL; growth media devoid of antibiotics); 2) CTL + Lipopolysac-

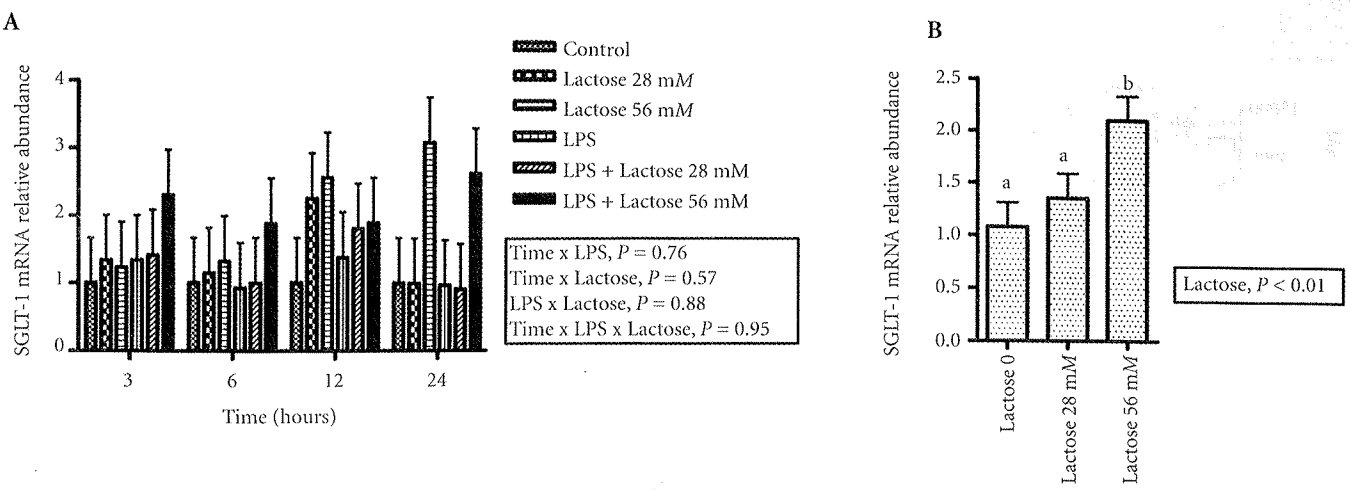

Figure 1. Effects of supplementation of crystalline lactose on relative abundance of SGLT-1 mRNA after 3, 6, 12, and 24 hours following treatment additions. Each bar represents the least-squares mean (± SEM) of three replications. Panel A represents the interactive time × LPS × lactose ($P = 0.95$) means. Panel B represents the main effects of lactose ($P < 0.01$).

charide (LPS; 10 ng/mL); 3) CTL + low lactose (28 m*M*); 4) CTL + high lactose (56 m*M*); 5) CTL + LPS + low lactose (28 m*M*); and 6) CTL + LPS + high lactose (56 m*M*).

Total RNA and cell culture media samples from both apical and basolateral compartments were harvested at 3, 6, 12, and 24 hours following the addition of treatments.

RNA Isolation and Quantitative Real-Time PCR Analysis

Total RNA was extracted and contaminating genomic DNA was removed from all RNA samples. Samples were reconstituted in nuclease-free water and frozen for further analysis. The quality of RNA was assessed by agarose gel electrophoresis and visulization of 28S and 18S rRNA bands. The quantity of RNA was determined by spectrophotometry (OD 260 nm). Complementary DNA (cDNA) was synthesized by reverse transcription (RT) from 1.0 μg of RNA. Reverse transcription reagents included 25 m*M* $MgCl_2$, 500 μ*M* dNTP's, 2.5 μ*M* random hexamers, 0.4 U/μL Rnase inhibitor, 50 U/μL MultiScribe reverse transcriptase, and TaqMan RT buffer. The RT mixture was incubated at 25°C for 10 min, heated to 37°C for 60 min, and inactivated at 95°C for 5 min. Synthesized cDNA was used as a template for real-time quantitative polymerase reaction (q-PCR) to quantify SGLT-1 mRNA relative to the quantity of the endogenous control (18S rRNA). The q-PCR reaction was conducted in 384-well plate with the SGLT-1 specific primers (TaqMan Gene Expression Assays, Applied Biosystems; Assay ID No. Ss03394307_m1). Commercially available eukaryotic 18S rRNA primers and a probe were used as an endogenous control. The PCR reactions, run in triplicate wells, were carried out with the Applied Biosystems 7900HT Fast Detection System using 40 cycles of amplification with alternating 15 sec, 95°C denaturation and 1 min, 60°C anneal/extension cycles.

Circulating Glucose Quantification

Media samples were used to quantify glucose concentration (Enzy-Chrom™ Glucose Assay Kit, Bioassay Systems) which were based on glucose oxidase and color reactions. The optical density of samples was read at OD 570 nm.

Statistical Analysis

Relative abundance of SGLT-1 mRNA in IPEC-J2 cells was calculated by ΔΔCT method using the average CT values of cells from control wells as the reference expression. These ΔΔCT values were analyzed using the MIXED procedure (SAS Inst. Inc., Cary, N.C.) to determine effects of SGLT-1 relative gene expression. The model consisted of the main effects of LPS, lactose, time, and their interactions. For glucose concentration quantification, the statistical model included effects of LPS, lactose, direction, time, and their interactions.

Results and Discussion

SGLT-1 mRNA Gene Expression in IPEC-J2 Cells

Relative abundance SGLT-1 mRNA of each treatment was shown in comparison with the control wells which have relative abundance SGLT-1 mRNA equal to one. There were no interactions of LPS × lactose × time (Figure 1A) or their two-way interactions. However, a main effect of lactose on increasing relative abundance of SGLT-1 mRNA ($P < 0.01$) in IPEC-J2 cells was detected when averaged across all time points (Figure 1B). Cells treated with a high concentration of lactose had greater SGLT-1 mRNA compared to the control ($P = 0.003$) and low lactose concentration ($P = 0.02$). This result agrees with previous research that reported feeding high glucose and galactose increased SGLT-1 mRNA abundance in mice. In addition, research also indicated that mRNA expression of SGLT-1 may be affected within hours, to one to three days after feeding a high-carbohydrate diet. Lastly, there were no differences

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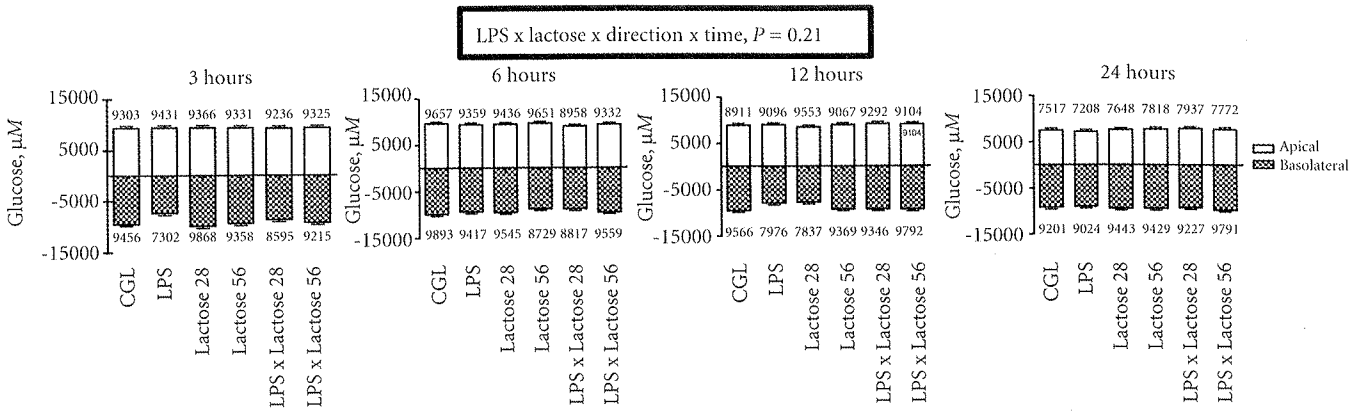

Figure 2. Effects of supplementation of low and high concentration of crystalline lactose on glucose concentration in IPEC-J2 cell culture media at 3, 6, 12, 24 hours following addition of treatments ($P = 0.21$). Each bar represents the least-squares means (± SEM) of three replications.

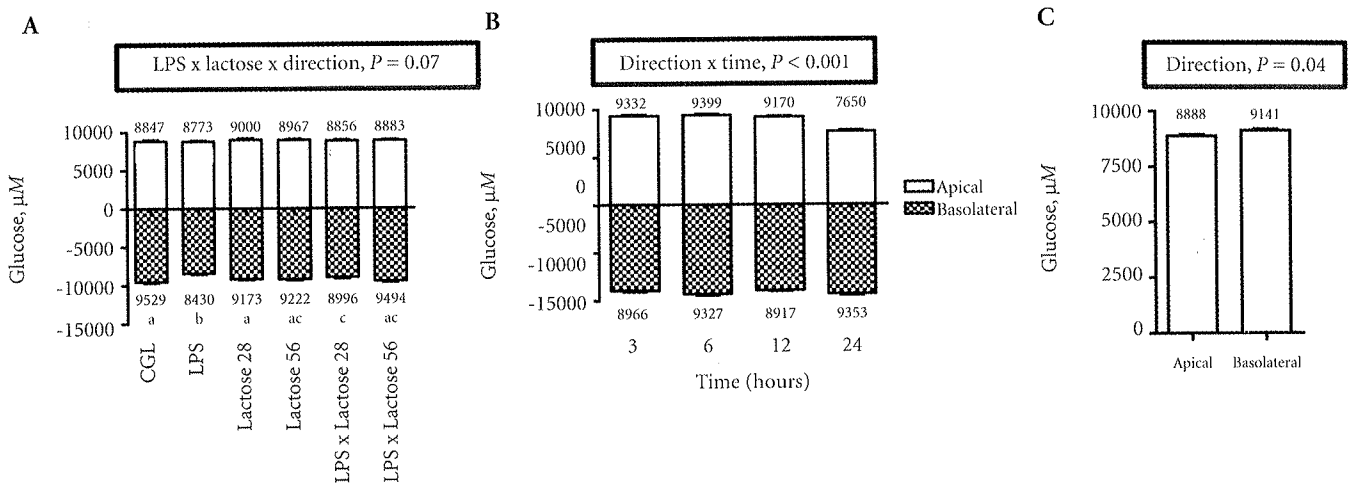

Figure 3. Effects of supplementation of low and high concentrations of crystalline lactose on glucose concentration in IPEC-J2 cells. Panel A represents interactive LPS × lactose × direction ($P = 0.07$) means. Panel B represents interactive direction x time ($P < 0.001$) means. Panel C represents polarized glucose transport (direction) among all timepoints and treatments ($P = 0.04$).

between control cells and cells treated with low lactose concentration on relative abundance of SGLT-1 mRNA.

Glucose Concentration in IPEC-J2 Cell Culture Media

Figures 2 and 3 illustrate the effects of supplementation of low and high concentration of crystalline lactose on glucose concentration in IPEC-J2 cell culture media at 3, 6, 12 and 24 hours following the addition of the respective treatments. There was no LPS × lactose × direction × time interaction (Figure 2). However, there was a LPS × lactose × direction interaction tendency ($P = 0.07$) when averaged across all time points ($P = 0.07$; Figure 3A). Lipopolysaccharide-treatment tended to inhibit glucose transport to the basolateral compartment compared to control wells. However, supplementation of lactose in LPS-treated cells tended to reduce ($P = 0.07$) the LPS-mediated inhibitory effect of glucose transport from the apical to the basolateral compartment.

As expected, glucose transport in IPEC-J2 was polarized basolaterally rather than apically (9,141vs. 8,888 μ*M*; $P = 0.04$; Figure 3C). The polarization of glucose transport basolaterally, followed a time-dependent pattern (direction × time, $P < 0.001$; Figure 3B). At 24 hours, glucose in the apical media (7,650 μ*M*) was significantly decreased compared to the basolateral (9,353 μ*M*) media and compared to 3, 6, and 12 hour apical media. In addition, glucose in the basolateral media at 24 hours was significantly greater compared to basolateral media at other timepoints.

Conclusions

This research indicates that crystalline lactose affects the expression of SGLT-1 mRNA, polarized glucose transport, and may restore LPS-induced reduction of basolateral glucose transport *in vitro* in model porcine jejunal epithelial cells.

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Effect of Dam Parity on Progeny Gastrointestinal Microbiota

The gastrointestinal microbiota of neonatal pigs may be affected by dam parity.

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Summary

Litter performance, progeny growth performance, and progeny health status may be affected by dam parity. The objective of the current experiment was to evaluate gastrointestinal microflora, as a measure of gut health, in progeny derived from first parity (P1) compared to fourth parity (P4) dams. Fecal samples were collected from the progeny (n = 6 pigs/litter) of P1 and P4 dams (n = 4 from each parity, P1 and P4) on days 1, 7, and 14 following parturition. Denaturing gradient gel electrophoresis was utilized to characterize gastrointestinal microbial populations and to calculate similarity and diversity indices. The similarity index represents the percentage of the microbial population that is similar within a group (P1 vs. P4). Diversity indices (Shannon's and Simpson's) represent the differences of the bacterial species within the microbial population. A greater Shannon's index and reduced Simpson's index are indicative of greater diversity among microbial populations. At all time points (days 1, 7 and 14), the fecal microbiota of progeny derived from P1 dams was more homogenous when compared to P4 progeny (P < 0.001). With respect to microbial diversity, P1 progeny tended (P = 0.07; Shannon's) to have greater microbial diversity compared to P4 progeny on day 1, and on day 7, the reduction in microbial diversity in P1 progeny reached statistical significance (Shannon's: P < 0.05). There were no differences in microbial diversity among progeny derived from different dam parities (P1 v. P4) on day 14. These results suggest that microbial populations, and thus health status, may be affected by dam parity.

Introduction

It is possible that progeny health status is affected by factors including (but not limited to) animal stress, passive immunity, and susceptibility to pathogens. When passive immunity is low or fails, the piglet's health status decreases and may affect survivability. Therefore, receiving adequate colostrum in the first 24 hours after birth is extremely important.

Preliminary observations reported in the *2008 Nebraska Swine Report* suggest that passive immunity, and thus health status, may be affected by dam parity. In order to substantiate our preliminary observations, another experiment was initiated and the results were published in the *2009 Nebraska Swine Report*. Parameters evaluated in the 2009 experiment included litter performance and transfer of passive immunity and the results can be summarized as follows: 1) No differences in litter performance were observed among first parity (P1) compared to fourth parity (P4) dams with the exception of litter birth weight which tended ($P = 0.10$) to be greater for P4 compared to P1 dams; 2) Immunoglobulin (Ig) A concentrations during lactation in colostrum and milk samples tended ($P = 0.08$) to be greater in samples collected from P4 compared to P1 dams; and 3) P4 progeny had greater ($P < 0.05$) serum IgG concentrations compared to P1 progeny throughout lactation. These results confirmed our preliminary observations that passive immunity may be affected by dam parity.

More information is needed to understand how dam parity may affect progeny health status. Recently, considerable evidence accrued that the composition of the intestinal microbiota of an individual may be linked and used as indicators of gastrointestinal health status. Therefore, factors that may affect establishment of the pig's gastrointestinal microbiota are likely to affect animal performance and include host physiology, environmental exposure, and diet.

Denaturing gradient gel electrophoresis (DGGE) is a technique that is capable of discriminating among bacterial species and is a means by which patterns of change in microbial populations can be detected through space and time (Thompson et al., 2008). Increase in microbial diversity has been associated with increased ecosystem stability and resistance to pathogen invasion (Konstantinov et al., 2004). In addition, species diversity affects a number of processes in ecological communities, including productivity, stability, and susceptibility to invasive species (Hooper et al., 2005). Therefore, the objective of the current experiment was to evaluate fecal bacterial population changes among P1 and P4 progeny as a means to further our knowledge on the effect of dam parity on progeny health status.

Materials and Methods

Experimental Design

The experimental protocol was reviewed and approved by the Institutional Animal Care and Use committee of the University of Nebraska–Lincoln. Dams (Large White × Landrace) utilized in the current study included P1

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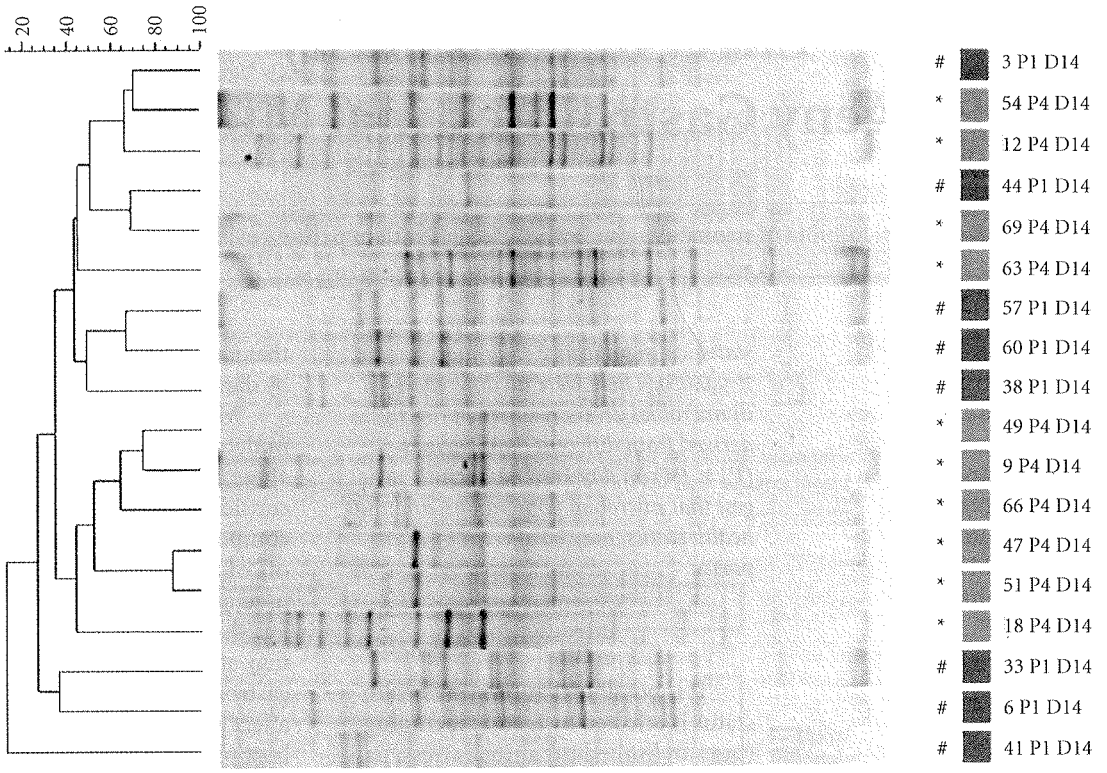


Figure 1. Dendogram derived from DGGE analysis of fecal bacterial community of piglets on day 14. UPGMA-type dendograms were constucted based on the similarity matrix resulting from Pearson's pair-wise comparisons of DGGE fingerprints. The red (#) squares represent P1 piglets and the green (*) squares represent P4 piglets.

gilts (n = 4) and P4 sows (n = 4). Dams were co-mingled and housed in stalls during gestation and moved to farrowing crates approximately five days prior to their expected farrowing date.

Fecal Sample Collection

Fecal samples were collected from six piglets from each litter (n = 4 from each dam parity, P1 and P4) on days 1, 7, and 14 following parturition. Fecal samples were stored in phosphate-buffered saline and frozen (-20°C) for further analyses.

Laboratory Analyses

Extraction of DNA from all fecal samples was carried out according to the methods described by Rasmussen et al. (2009). The resultant DNA was utilized for subsequent polymerase chain reaction (PCR) and DGGE analyses. Briefly, for investigation of the entire microbe population in fecal samples, PCR was performed by using universal primers to amplify the V3 region of the 16S rRNA gene. Denaturing gradient gel electrophoresis was performed as described by Walter et al. (2000). Denaturing gradient gel electrophoresis images were analyzed using BioNumerics software where the DGGE fingerprints were transformed to peak profiles and intensities of individual bands were determined as a percent peak surface area relative to the surface area of the entire molecular fingerprint of the sample. To determine the effect of dam parity, normalized fragment intensities of all bands in DGGE fingerprints were determined and compared among dam parities, which is partially depicted by the dendogram in Figure 1.

To determine the microbial diversity of the fecal DNA samples, Shannon's and Simpson's ecological indices were applied to the molecular fingerprints as described by Scanlan et al. (2006). Briefly, Shannon's diversity index was calculated using the formula shown below in which *pi* represents the proportions of a species *i* present in a sample (determined as the proportion of the band intensity with respect to the intensity of the entire fingerprint) of *n* different species (number of bands in the profile). Simpson's diversity index was calculated with the following formula in which *ni* represent the number of organisms belonging to species *i* (determined as proportion of the band intensity with respect to the intensity of the entire fingerprint) and *N*, the total number of organisms in the microbial population.

$$\text{Shannon's index} = \sum_{i=1}^{n} -pi * Ln\,(pi)$$

$$\text{Shannon's index} = \sum_{i=1}^{n} \frac{-ni*(ni-1)}{N(N-1)}$$

Statistical Analysis

The GLM procedure (SAS Inst. Inc., Cary, N.C.) was used to analyze all parameters as a completely random design with repeated measures over time on each experimental unit. The model included terms for the fixed effects of parity and time and their interaction. Comparisons among dam parity and time were made only when a significant ($P < 0.05$ unless noted otherwise) F-test for the main effect or interaction was detected using the least significant dif-

Table 1. Similarity indices[a] of microbial populations in piglets (n=6).

	Parity 1[b]	Parity 4
Day 1	66.31 ± 3.12	34.74 ± 3.12
Day 7	55.69 ± 3.12	33.81 ± 5.93
Day 14	48.56 ± 3.12	33.42 ± 3.12

[a]Similarity indexes are calculated using the BioNumerics software, the DGGE fingerprints were transformed to peak profiles. Intensities of individual bands were determined as a percent peak surface area relative to the surface area of the entire molecular fingerprint of the sample.
[b]Parity x day $P < 0.05$.

Table 2. Diversity indices[a] of microbial populations in piglets (n=6).

	Parity 1		Parity 4		*P*-value	
	Shannons	Simpsons	Shannons	Simpsons	Shannons	Simpsons
Day 1	1.85 ± 0.21	0.30 ± 0.09	1.20 ± 0.21	0.37 ± 0.09	0.03	0.60
Day 7	2.80 ± 0.21	0.08 ± 0.09	2.30 ± 0.23	0.16 ± 0.10	0.12	0.40
Day14	2.65 ± 0.21	0.09 ± 0.09	2.43 ± 0.21	0.13 ± 0.09	0.46	0.73

[a]Diversity Indexes were calculated by comparing molecular fingerprints of DNA. A higher Shannon's diversity index represents more diversity. A lower Simpson's diversity index represents more diversity.

ference procedure. All means presented are least-squares means.

Results and Discussion

Denaturing gradient gel electrophoresis analysis revealed that there was substantial variation in gut microbiota composition among individual piglets (Figure 1). Similarity indexes for fecal microbial populations are depicted in Table 1. These coefficients represent the similarity of the microbial population within a group (P1 or P4). This analysis revealed that on days 1, 7, and 14, the microbial population of P1 piglets was more uniform when compared to P4 progeny ($P < 0.001$). Diversity indices (Shannon's and Simpson's) represent the differences of the bacterial species within the microbial population while each index weighs species richness and evenness slightly differently. Shannon's index incorporates species richness (number of species, or in this case, PCR-DGGE bands) and evenness (the relative distribution of species) and Simpson's index takes into account the number of species present, as well as the relative abundance of each species. An increasing Shannon's index signifies a more diverse microbial population, while a decreasing Simpson's index indicates a greater diversity. Differences in microbial populations with respect to microbial diversity using the Shannon's and Simpson's indices were determined and are represented in Table 2. Shannon's microbial diversity index was greater ($P < 0.03$) for P1 progeny on day 1 compared to P4 progeny, indicating that P1 piglets have greater microbial diversity compared to P4 progeny. There were no differences among parity found in Simpson's diversity index.

Collectively, differences in similarity indicate the presence of different bands (i.e., bacterial species) and differences in microbial diversity indicate an overall change in microbial community complexity. Therefore, with respect to P4 progeny, P1 progeny have greater similarity (i.e., fewer bacterial species) throughout the preweaning phase, but have greater diversity (i.e., number of bacterial species present and their relative abundance) through day 7 preweaning.

There are several factors which may account for the differences in gut microbial ecology that were observed in P1 and P4 progeny. Most importantly, the composition of sow milk may affect the bacterial population of its progeny. We have previously observed a numerically greater concentration of IgA in the colostrum and milk of P4 sows (*2009 Nebraska Swine Report*) and it is possible that this difference in immunoglobulins could account for some of the differences observed in microbial populations among progeny derived from different parities. Secretory IgA in colostrum is an important factor of microbial control in the piglet intestinal tract (e.g., control of pathogenic bacteria), as the piglets themselves have not yet developed a functional immune system. Therefore, IgA in milk is likely to affect the microbiota that become established in the gastrointestinal tract of the piglets. The greater IgA concentrations in P4 piglets would exert a greater selective force and could result in the lower microbial diversity in the gut of P4 progeny that we have observed in this study.

With respect to changes in the bacterial population (similarity and diversity), over time these changes could affect the functions that the microbial community supplies to the host and how the host responds to these changes. For example, changes in the microbial community could shift the production of short-chain fatty acids (i.e., butyrate) that may have an anti-inflammatory effect on the gut. Alternatively, changes in gut microbiota may affect the way in which the host responds to different microbes immunologically. That is, whether the host tolerates (immune response not initiated) or responds (immune response initiated with resultant inflammatory events) to changes in microbial populations.

Conclusion

Results from this experiment indicate that there are differences in microbial populations among progeny derived from different dam parities. However, more research is needed to determine how these changes may affect health status and growth performance.

[1]Erin E. Hinkle, Huyen Tran, and Justin W. Bundy, graduate students; Roman Moreno, graduate student and research technologist; Phillip S. Miller, professor; Duane E. Reese, associate professor and extension swine specialist; and Thomas E. Burkey, assistant professor, Animal Science Department, University of Nebraska–Lincoln. Jens Walter, assistant professor, Food Science Department, UNL. Matthew W. Anderson, manager, and Jeffrey M. Perkins, research technician at the UNL Swine Research Farm.

Effect of DNA Markers in Nebraska Selection Lines

Rodger Johnson[1]

Summary

DNA from 57 generation-28 boars that had sired progeny in the NE selection and control lines was submitted to GeneSeek Inc., where genotypes for eight Single Nucleotide Polymorphic Markers (SNPs) affecting economic traits in pigs were determined. Three markers are reported to be associated with growth and composition of growth, three with meat quality, and two with number of live pigs per litter. Frequencies of marker alleles were estimated in two selection lines and in their respective controls to determine whether selection had increased the frequencies of alleles associated with increased performance. Relationships of boar marker genotype with growth, backfat, and loin eye area were studied by regressing both boar phenotype and progeny phenotype on the number of favorable alleles in the boar's genotype. Frequencies of markers affecting reproduction (ESR and EPOR) were inconsistent with the selection background of the lines. Frequencies of alleles of CCKAR and MC4R, markers that affect growth and composition of growth, in selection and control lines are consistent with observed selection responses, suggesting that the allele that decreased backfat was being selected for. Regression analyses were consistent with that result. There was little evidence there had been selection for meat quality markers in these lines. The study demonstrated that selection for markers in some populations may not produce desired responses.

Introduction

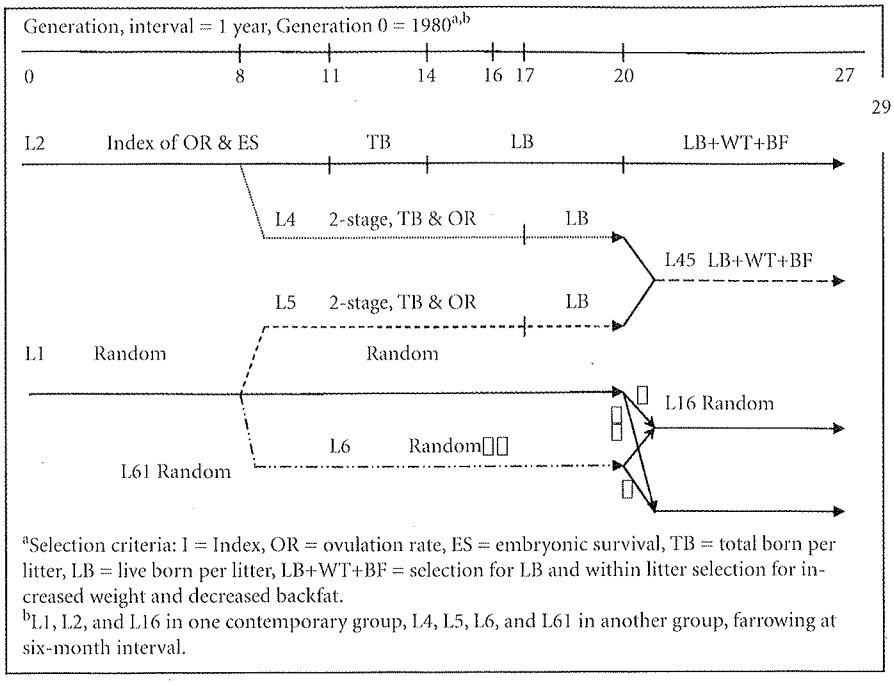
A large number of genetic markers associated with economic traits in pigs have been identified. But for several reasons, relatively few of them are being used to enhance response to selection in commercial populations. Genes are DNA sequences within chromosomes that contain the code (order of nucleotides) to produce a specific protein. Markers are not the entire gene but rather are very small segments of the chromosome where differences among individuals can be identified. There are many different types of markers, but most markers used today are **single-nucleotide polymorphism** (**SNP**, pronounced *snip*) which is a DNA sequence variation occurring when a single nucleotide – A, T, C, or G – differs among individuals or between the paired chromosomes of an individual. For example, two sequenced DNA fragments from different individuals, AAGCCTA to AAGCTTA, contain a difference in a single nucleotide. Thus, there are two alleles (C and T) for this marker.

Some markers are within the coding region of a gene with a causative effect on an economic trait. But most markers are not in coding regions of causative genes but are on the same chromosome positioned close to a causative gene. In those cases, the gene and the marker are linked and they tend to be inherited together. Then, marker genotype tells us something about whether the individual contains a desirable copy of the causative gene. Thus, the value of a marker depends on the linkage relationship between the causative gene and the marker. Markers loosely linked with causative genes are of limited value. Even when closely linked, which marker allele is linked with the desirable allele of the causative gene may differ among populations. As a result, selection for a particular marker allele to enhance response in an economic trait may be effective in one population, but ineffective in another.

Even if markers are within causative genes, the effectiveness of selecting on them may differ among populations because average gene effects in the population are frequency dependent. Genes have their greatest average effect and selection for desirable alleles produces the greatest response when alternative alleles (different forms of the gene) have intermediate frequencies, between 0.25 and 0.75. If the better allele is at high frequency, then little extra increase in performance from pushing its frequency even closer to one is available. When the better allele has low frequency, it is rare and variation at that gene locus may explain very little of the genetic variation in the trait. However, long-term selection opportunities are greatest when initial frequency of desirable alleles is low. Even when alleles of causative genes have intermediate frequencies, their effects may be relatively small in proportion to the total genetic variation for the trait, and selecting on these markers may cause only small changes in performance. Thus, many questions about which markers to use and their value in selection programs still exist.

Long-term selection in pigs at the University of Nebraska for increased reproduction, increased growth, and decreased backfat has produced lines that differ from randomly selected control lines by more than 50% in litter size and 12 to 15% in rate of growth and backfat thickness. Frequencies of marker alleles are expected to differ between selection and control lines if genetic markers are associated with these traits. Previous research identified more than 30 regions of the chromosomes that harbor genes affecting both reproduction and growth traits in these lines, but positions of causative genes were not identified precisely enough (close linkage was not established) to use these markers in selection.

A few markers in the pig genome have been researched in great depth, and there is a high degree of confidence in their effects on the discovery populations. These markers are either within the DNA sequence of causative genes or very tightly linked with causative genes. GeneSeek Inc., Lincoln, Neb., provides genotyping services for eight markers whose effects on reproduction, growth, or pork quality are estimated quite precisely. None



[a]Selection criteria: I = Index, OR = ovulation rate, ES = embryonic survival, TB = total born per litter, LB = live born per litter, LB+WT+BF = selection for LB and within litter selection for increased weight and decreased backfat.
[b]L1, L2, and L16 in one contemporary group, L4, L5, L6, and L61 in another group, farrowing at six-month interval.

Figure 1. Evolution of the Nebraska selection lines.

of these markers were identified in Nebraska selection lines. The purpose of this report is to estimate allele frequency and marker effects in the Nebraska selection lines for the panel of genes for which GeneSeek Inc. provides commercial genotyping services.

Methods

The Nebraska lines include a selection and control line in each of a summer (Lines 2 and 16) and winter (Lines 45 and 61) farrowing group. All lines derived from the same base population, a Large White x Landrace cross made in 1979. Evolution of the lines is illustrated in Figure 1. Line 2 was selected 11 generations for an index of ovulation rate and embryonic survival, nine generations for increased total born or live born per litter, and nine generations for increased live born per litter, increased 180-day weight, and decreased backfat thickness. Line 1, the control line in the summer group through generation 20, was selected randomly. Three additional lines (Lines 4, 5, and 6), derived from Lines 1 and 2, were formed in Generation 8 and made up the winter group. Line 4, derived from Line 2, and Line 5, derived from Line 1, were selected eight generations for ovulation rate and total born per litter and then three generations for live born per litter. Lines 4 and 5 were then crossed to form Line 45 which has been selected for nine additional generations for increased live born per litter, increased 180-dayweight, and decreased backfat thickness in the same way as Line 2 was selected during that time. Line 6 was selected randomly from Generations 8 to 20. At Generation 20, control Lines 1 and 6 were crossed to from Lines 16 and 61, which were each continued with random selection. Thus, Lines 2 and 45 have undergone 29 generations of selection for increased litter size with added selection for increased growth and decreased backfat in the last nine generations.

Generation interval for all lines has been one year as only gilts were farrowed. Line sizes were 40 to 80 litters per generation by 14 to 20 sires. Selection rates during all generations have been 1/4 to 1/3 for females and 1/6 to 1/4, depending on the selection criteria, for males.

Tissue samples collected from all breeding boars of generation 29 were submitted to GeneSeek, Inc. and their genotypes for eight markers were determined. The boars were considered to adequately represent the population. They contribute one-half of the genes to the progeny generation, and most of them also have full and half sibs that were selected. Gene frequencies of female parents in this generation are expected to be similar to that of the boars.

Gene Marker Descriptions and Favorable Alleles

Information about the gene markers evaluated was obtained from the GeneSeek, Inc. Web site (*www.geneseek.com/prod_pigs.php*). Two of the gene markers (**ESR** and **EPOR**) have been reported to have significant effects on litter size, operating primarily on uterine capacity or embryonic survival. If these genes have contributed significantly to variation in litter size, then frequencies of favorable alleles are expected to be higher in selection lines than in controls. Three of the gene markers (**CCKAR, HMGA1,** and **MC4R**) affect growth and/or composition of growth and selection during the last nine generations is expected to have changed frequencies of their alleles. Three of the gene markers (**CAST249, CAST 638**, and **PRKAG3**) affect meat quality and are not known to affect any of the traits selected for in Lines 2 and 45. More information regarding these markers is presented in the appendix of this paper.

Estimation of Marker Effects

Frequencies of marker alleles were determined from the distributions of genotypes in each line. Effects of the genes for growth and meat quality were estimated with regression. First, the boar's own phenotype was regressed on the number of favorable alleles in the boar's genotype, which estimates the average increase or decrease in boar performance per copy of the favorable allele. A total of 57 boars were selected as breeders, 11 to 16 per line, too few for highly reliable estimates of marker effects; thus, these regressions have quite large standard errors. The relationship between sire marker genotype and progeny phenotype was also estimated by regressing

(Continued on next page)

sire's progeny phenotype on the number of copies of the favorable allele in sire's genotype. Sire progeny phenotype provides an estimate of one-half of the sire's breeding value. Each boar had between 10 and 25 progeny, so these regressions are somewhat analogous to regressing 1/2 sires breeding value on his marker genotype. The number of progeny was insufficient for a highly accurate estimate of each boar's breeding value, but averaged across all boars, this method provides quite reliable estimates of marker effects. Regression analyses could not be done for the reproduction markers, **ESR** and **EPOR**, as daughters of these boars have not yet produced litters.

Results

Litter size means for Generation 29 dams and growth trait means for Generation 30 progeny are in Table 1. The two selection lines (Lines 2 and 45) differ from respective controls by 37 to 48% in live pigs per litter, 11 to 12% in 180-day bodyweight, and -12 to -14% in backfat thickness. Selection has not caused change in longissimus muscle area.

Genotypic distributions and allele frequencies of Generation 29 sires are in Table 2. Frequencies of alleles are presented as the probability of the favorable allele (e.g., Pr (A)) for each gene.

Reproductive Genes. The ESR gene marker was not segregating in either of the selection lines — the frequency of the favorable allele was zero. Only one copy of the favorable ESR allele existed in this sample of boars and it was in a control, Line 16 boar. This same ESR polymorphism was genotyped in Lines 4, 5, and 6 at Generation 16. At that time, the frequency of the favorable G allele was .06 in Line 4 and 0 in Lines 5 and 6. Thus, the ESR polymorphism was segregating in the base population but probably at low frequency. It is a marker for litter size, not a causative gene, and linkage relationships were different in this population from the ones in which the marker allele was discovered and had an effect. There clearly has not been selection on the ESR marker in the selection lines.

Table 1. Means for generations 29 (litter traits) and 30 (growth traits).

	Line 2		Line 16		Line 45		Line 61	
Trait	n	Mean	n	Mean	n	Mean	n	Mean
Total Born	32	13.3	41	8.9	36	16.1	37	10.1
Live Born	32	11.5	41	8.4	36	14.1	37	9.5
180-day Wt, kg	195	103.2	94	92.2	219	104.8	87	94.5
10 rib backfat, cm	195	2.05	94	2.38	219	2.12	87	2.42
Longissimus area, cm^2	195	28.2	94	28.6	219	28.7	87	28.2

The other gene with reported effects on litter size, EPOR, was segregating in all lines, but the frequency of the favorable T allele was very low in both Lines 2 and 45 compared with Control Lines 16 and 61. It has been reported that females with two copies of the T allele (genotype TT) have approximately one more pig per litter than those homozygous for the C allele (genotype CC). If that relationship existed in these populations, it is highly likely that the frequency of the T allele would be much greater in both selection lines, especially as compared with the control lines. Either the EPOR polymorphism is not a causative gene, but is linked with another gene affecting litter size, or its effect is less in this population than in others so that it explains only a small proportion of the variation in litter size. Whichever the case, it is unlikely that selection on the EPOR polymorphism will enhance response to selection in these lines.

Growth and Carcass Genes. The **CCKAR** marker is associated with feed intake and growth. The frequency of the favorable G allele was low in Line 2 relative to Line 16 (.23 vs. .91), and high in both Lines 45 and 61 (.72 vs. .90). Although it is not reported that the gene affects backfat, greater feed intake often causes increased backfat. The lower frequencies in both selection lines, relative to respective controls, may be the result of selection for leanness. Results for the **MC4R** polymorphism are consistent with that relationship. The A allele of **MC4R** causes pigs to grow faster and the G allele causes them to be leaner. The frequency of the A allele was intermediate in both control lines (Lines 16 and 61) and low in selection lines (Lines 2 and 45). There was selection for both growth and leanness in the selection lines. Increased frequency of the allele conferring leanness rather than the one for growth indicates that selection for lean placed more weight on this locus than did selection for growth if the marker associations are the same in the Nebraska lines as in the discovery populations. Allele frequencies for **HMGA** are intermediate in all lines and appear to not have been affected greatly by selection.

Meat Quality Genes. There is no reason to believe that frequencies of alleles for the three markers with effects on meat quality (**CAST249, CAST 638**, and **PRKAG3**) should have been changed by selection as none of these markers have been reported to affect reproduction, growth, or carcass traits. The frequencies in these lines are of interest simply to characterize changes not expected to be related to selection. All lines had intermediate frequencies of Cast 249 and are not greatly differentiated. Line differences could easily be the result of random genetic drift. All lines, except Control Line 61, had high frequencies of the favorable A allele of CAST 638. It is likely that the frequency of this allele was relatively high in the base generation and has drifted down in Control Line 61, or it was at intermediate frequency in the base generation and drifted up in Lines 2, 45, and 16. In either case, there is some opportunity to improve meat quality in the selection lines by selecting for the AA genotype of CAST 249. Because there is already a high frequency of the AA genotype for CAST 638, little additional response is expected from selecting for the AA/AA haplotype of the two markers.

The **PRKAG3** marker was not segregating in Line 2, for which the

Table 2. **Genotypes of Generation 29 sires in lines selected for litter size, growth and backfat (Lines 2 and 45) and respective controls (Lines 16 & 61). See text for description of genes and favorable allele.**[a]

CAST_249		CAST_638		CCKAR		EPOR		HMGA		MC4R		ESR		PRKAG3	
Genotype	N	Genotype	N	Genotype	N	Genotype	N	Genotype	N	Genotype	N	Genotype	N	Genotype	N
Line 2															
AA	4	AA	15	AA	8	CC	13	CC	3	AA	0	AA	15	AA	0
AG	8	AC	0	AG	7	CT	2	CT	8	AG	0	AG	0	AG	0
GG	3	CC	0	GG	0	TT	0	TT	4	GG	15	GG	0	GG	15
Pr(A)		Pr(A)		Pr(G)		Pr(T)		Pr(T)		Pr(A)		Pr(G)		Pr(A)	
0.53		1.00		0.23		0.07		0.53		0.00		0.00		0.00	
Line 16															
AA	1	AA	8	AA	0	CC	6	CC	2	AA	3	AA	10	AA	3
AG	7	AC	3	AG	2	CT	4	CT	7	AG	6	AG	1	AG	4
GG	3	CC	0	GG	9	TT	1	TT	2	GG	2	GG	0	GG	4
Pr(A)		Pr(A)		Pr(G)		Pr(T)		Pr(T)		Pr(A)		Pr(G)		Pr(A)	
0.41		0.86		0.91		0.27		0.50		0.55		0.05		0.45	
Line 45															
AA	1	AA	13	AA	0	CC	10	CC	5	AA	0	AA	16	AA	4
AG	6	AC	3	AG	9	CT	6	CT	10	AG	4	AG	0	AG	8
GG	9	CC	0	GG	7	TT	0	TT	1	GG	12	GG	0	GG	4
Pr(A)		Pr(A)		Pr(G)		Pr(T)		Pr(T)		Pr(A)		Pr(G)		Pr(A)	
0.25		0.91		0.72		0.19		0.37		0.13		0.00		0.50	
Line 61															
AA	2	AA	4	AA	0	CC	6	CC	9	AA	1	AA	15	AA	3
AG	5	AC	6	AG	3	CT	6	CT	5	AG	8	AG		AG	6
GG	8	CC	5	GG	12	TT	3	TT	1	GG	6	GG	0	GG	6
Pr(A)		Pr(A)		Pr(G)		Pr(T)		Pr(T)		Pr(A)		Pr(G)		Pr(A)	
0.30		0.47		0.90		0.40		0.23		0.33		0.00		0.40	

[a]Pr = probability of the favorable allele.

frequency of the favorable allele was 0, and alleles had intermediate frequencies in other lines. It is likely that allele frequencies were intermediate in the base population and random drift, not selection, caused the favorable allele to be removed from Line 2, assuming the frequency was zero in dams as well.

Regressions. Regression coefficients are in Table 3. The most reliable ones are for progeny phenotype on sire genotype. The G allele of **CCKA R** is associated with increased feed intake and growth. Its effects in this sample were inconsistent, being positive for boar 180-day weight (4.87 ± 2.06 kg per copy), but negative for progeny weight (-1.88 ± 1.00 kg per copy). It was significantly associated with decreased LEA in progeny, but not boar LEA, and did not affect backfat.

Estimates of the effects of the T allele of **HMGA** were consistent in both boar and progeny. Each additional copy was associated with increased 180-day weight, (1.97 ± 1.57 and 1.23 ± .75 kg), decreased backfat (-.13 ± .065 and -.056 ± .026 cm per copy) and decreased LEA, (-1.57 ± 0.56 and -.64 ± .23 cm^2 per copy in boars and progeny, respectively).

The **MC4R** marker is known to be within the causative gene as the effect of this marker is consistent across many populations and results here are in agreement. Each copy of the A allele was associated with increased boar 180-day weight (4.05 ± 1.96 kg) and increased progeny weight (4.07 ± 1.03 kg). The A allele also significantly increased progeny backfat (0.08 ± 0.036 cm per copy). These results are consistent with changes in allele frequencies in which selection in Lines 2 and 45 was for the allele that conferred greater leanness.

There was some evidence that the meat quality genes (**CAST249**, **CAST638**, and **PRKAG3**) also affected growth and leanness. Regressions of progeny phenotype on number of copies of the favorable allele were significant for **CAST 249** (backfat), **CAST 638** (LEA), and **PRKAG3** (backfat). Progeny 180-day weight increased 2.65 ± 0.72 kg with each copy of the **CAST 249** A allele in sire's genotype. Progeny LEA increased 0.72 ± 0.35 cm^2 for each copy of the A allele of **CAST 638**, and progeny backfat decreased 0.068 ± 0.027 cm with each copy of the A allele of **PRKAG3**. In each case, regressions of boar's phenotype on number of copies of the favorable allele in the boar's genotype produced regressions with the same sign, although they were not significant, lending additional evidence that these genes affected these performance traits. However, these genes probably explain only a small percentage of the variation in these traits and were under weak selection as changes in allele frequencies (Table 2) are either small or inconsistent with regression results.

Discussion

This study demonstrates why analyzing marker genotypes in small selection lines may not tell much about whether significant selection has been applied to individual loci. Results are often inconsistent with expectations. Part of the explanation is that studies to identify important candidate genes

(Continued on next page)

Table 3. Regressions of phenotype on number of favorable alleles (b), standard errors of regressions (se), and probability regressions differ from zero (p).

	Regressions of boar's own phenotype on number of favorable alleles in boar's genotype			Regressions of boar's progeny phenotype on number of favorable alleles in sire's genotype		
Trait	b	se	p	b	se	p
			CCKAR			
WT, kg	4.87	2.06	0.02	-1.88	1.00	0.06
BF, cm	0.076	0.094	0.42	-0.006	0.035	0.86
LEA, cm^2	-0.098	0.83	0.91	-0.82	0.31	0.008
			HMGA			
WT, kg	1.97	1.57	0.21	1.23	0.75	0.1
BF, cm	-0.13	0.065	0.04	-0.046	0.026	0.07
LEA, cm^2	-1.57	0.56	0.007	-0.64	0.23	0.005
			MC4R			
WT, kg	4.05	1.96	0.04	4.07	1.03	0.0001
BF, cm	-0.007	0.09	0.93	0.08	0.036	0.03
LEA, cm^2	-0.13	0.78	0.87	0.26	0.32	0.42
			CAST249			
WT, kg	1.84	1.51	0.23	2.65	0.72	0.0003
BF, cm	-0.096	0.062	0.13	0.012	0.026	0.64
LEA, cm^2	-0.305	0.56	0.59	-0.23	0.23	0.31
			CAST638			
WT, kg	-1.48	2.01	0.47	-1.53	1.14	0.18
BF, cm	-0.22	0.08	0.01	-0.026	0.04	0.51
LEA, cm^2	0.46	0.76	0.55	0.72	0.35	0.04
			PRKAG3			
WT, kg	2.97	1.49	0.05	0.23	0.78	0.76
BF, cm	-0.035	0.066	0.59	-0.068	0.027	0.01
LEA, cm^2	0.26	0.58	0.65	-0.41	0.24	0.08

usually report differences between extremes. If the marker is an A/G polymorphism, mean phenotypes for individuals with AA and GG genotypes are estimated (AA – GG) or the mean phenotype for heterozygotes compared with the mean of the homozygotes (AG – ½ (AA + GG)) is estimated. For example, the difference between TT and CC genotypes at **EPOR** has been estimated at one pig per litter. But the effect of the T allele in a selection line and the selection applied to it, relative to other genes affecting litter size, are frequency dependent. In fact, they are approximately equal to the ratio of genetic variance at that locus relative to total genetic variance for the trait. That ratio decreases as frequency of T increases. Thus, when frequency of an allele with big effects, as estimated by difference between homozygotes gets up to .5 or greater, there is increasingly less selection on it. In fact there may be very little selection applied at that locus relative to all the other genes influencing the trait. Then, genetic drift is the most powerful force influencing allele frequencies.

Genotyping for a small number of markers and then practicing selecting mainly or only on marker genotypes can be a large mistake. A better approach is to include the markers in estimating breeding values because that method accounts for marker frequencies if marker genotypes are known for all selection candidates and produces the most accurate estimates of breeding values. When allele frequencies get to intermediate values, there may be little change in frequency of a gene with fairly large effect. In larger commercial populations, allele frequencies are expected to be at values that optimize response to selection. If drift moves the frequency of the desired allele down, then in the next generation there will be a bit more pressure on that allele and it will move back up. If by chance, in some generation both drift and selection move the frequency higher, then in the next generation there will be even less selection on that locus. After a very large number of generations and without mutation, fixation of favorable alleles can occur. But if populations are small, fixation of the undesirable allele also can occur. Because many of the reported markers with effects on economic traits are really linked markers and linkage relationships are different across populations, there can be considerable variation in marker effects across populations. Breeders are advised to not select on individual marker genotypes, but if genotypes are known on all candidates for selection, include the data in the EBV process.

[1]Rodger Johnson, professor, Animal Science Department, University of Nebraska–Lincoln.

Appendix

Description of Gene Markers (www.geneseek.com/prod_pigs.php)

CAST* (U.S. Patent Application #20,070,172,848): Calpastatin (CAST) is a specific inhibitor of μ- and m-calpain proteases. There is evidence indicating that in different species, including the pig, calpastatin activity post-mortem is highly related to meat tenderness. Two missense mutations (**CAST *0188I*** or ***Arg249Lys*** and **CAST PvuII** or ***Arg638Ser***) were identified and when used in tandem, are significantly associated with firmness, juiciness, Instron force, chewiness, and tenderness scores. Both mutations can be genotyped and used individually. The A allele is the favorable allele for the first mutation (**CAST *249Arg* (SNP=A)**) and is associated with higher tenderness, lower cooking loss, and Instron force.

Similar effects were observed with the second *CAST* mutation: ***CAST Arg638Ser.*** This mutation was also found to be a significant source of variation for cured ham moisture content. The A allele of ***CAST 638Arg (SNP=A)*** is again the favorable allele and is associated with higher moisture in the cured ham than ***CAST 638Ser (C allele).***

Both mutations can be used together as a haplotype maximizing the accuracy of selection for tenderness, cooking loss, and related traits. Haplotype **249Lys/Arg638** is the favorable haplotype (SNP's A/A).

(Ciobanu et.al., J Anim Sci. 2004 Oct;82(10):2829-39.)

CCKAR: The cholecystokinin type A receptor (CCKAR) genetic test is associated with physiological control of feed intake, hunger fulfillment, and obesity. Animals with at least one copy of the dominant G-variant have, on average, ~5% greater daily feed intake, 3% greater daily gain, and 3% fewer days to reach 180 kg, when compared to homozygotes for the A-variant.

SNP G = Favorable allele for growth

(Houston et. at., Genetics. 2006 Nov; 174(3):1555-63.)

Erythropoietin (EPOR): A genetic variant in the swine erythropoietin receptor gene is associated with uterine capacity and litter size. The favorable genetic variate (T allele) has demonstrated an increase in uterine capacity as well as an increase in live births in two different swine populations at USDA-MARC. In a commercial herd, an extra pig per litter was observed when comparing boars that have two copies of the favorable **EPOR** marker (TT) versus boars with zero copies (CC). The T allele is the favorable allele.

(Vallet, J.L., et. al., Animal Genetics. 2005 36(2): 97-103).

HMGA1* (U.S. Patent No. 20,040,029,145) : The high mobility group AT-hook protein 1 (HMGA1) genetic test is associated with lean mass percentage, growth and backfat in several swine breeds. The T allele is favorable (T-variant at position 576) and pigs with that allele are likely to be leaner and produce offspring that are leaner than those with the C allele.

(Kim *et. al.*, Obes Res. 2004 Dec;12(12):1981-94.)

MC4R* (U.S. Patent #6,803,190): The melanocortin-4 receptor (**MC4R**) is expressed in virtually all brain regions of mammals and plays an important role in energy homeostasis. **MC4R** has been described in several studies as a functional gene controlling several growth and performance traits in pigs. Allele frequencies of a polymorphism (Asp298Asn) were quite different among commercial pig breeds where divergent selection has been practiced intensively. In general, Asn298 allele (SNP=A) is associated with higher average daily gain and backfat thickness. Conversely, the Asp298 allele (SNP=G) is associated with lean growth with high feed conversion rate.

Allele (SNP) A = (Asn298-ASPARAGINE): Pigs with genotype A/A grow significantly faster (37 g/day) and consume more daily feed (~8%) than pigs that are are G/G. Allele (SNP) G: = (Asp298-ASPARTIC ACID): Pigs that are G/G have 9% less backfat and lower feed intake than pigs that are A/A. The allele effects appear to be additive. The heterozygotes fall between the two homogygotes.

(Kim et. al., Mamm Genome. 2000 Feb;11(2):131-5.)

Estrogen Receptor (ESR) U.S. Patent #5,550,024: Estrogen plays an essential role in several reproductive functions, including expression of estrus, fertility, embryo and fetal development, and maintenance of pregnancy. A genetic variant of the ESR gene (allele G) is associated with increased litter size. Females that carry one copy of the favorable variation of the gene (G-SNP) will, on average, yield 0.4 pigs per litter increase. Homozygotes (2 copies, GG) for this genetic variation yield 0.8 pigs per litter increase (average) compared with those homozygous for the A allele (AA). This test is reported to be effective in Large White or Yorkshire breeds or crosses that involve them. The G allele is favorable.

Rothschild, et. al., Proc. Natl. Acad. Sci. 1996 Jan; Vol. 93: 201-205

PRKAG3* (U.S. Patent #6,919,177): PRKAG3 is a regulatory subunit of AMP-activated protein kinase, which is involved in the regulation of energy homeostasis in eukaryotes. The *PRKAG3* gene is well known for one of its alleles called RN– (200Q), present only in Hampshire pigs. This mutation affects glycogen content in muscle and, in general, meat quality traits of pigs that include ultimate pH and color measures which are correlated with other characteristics like drip loss, water holding capacity, tenderness, and cooking loss. Another mutation, I199V, which is nearby and causative as well, affects also glycogen content, ultimate pH and color, but this mutation is present in all breeds. The favorable allele is 199I (SNP=A) and is associated with lower glycogen, higher ultimate pH and favorable color. The differences between homozygotes account for .1 ultimate pH between I/I (SNP= A/A) and V/V (SNP G/G) animals with the heterozygotes being intermediate. In addition, the I/I animals are significantly better for lower glycolytic potential, better color and Minolta reflectance scores. SNP A = Isoleucine (I), the favorable allele SNP G = Valine (V) = unfavorable allele

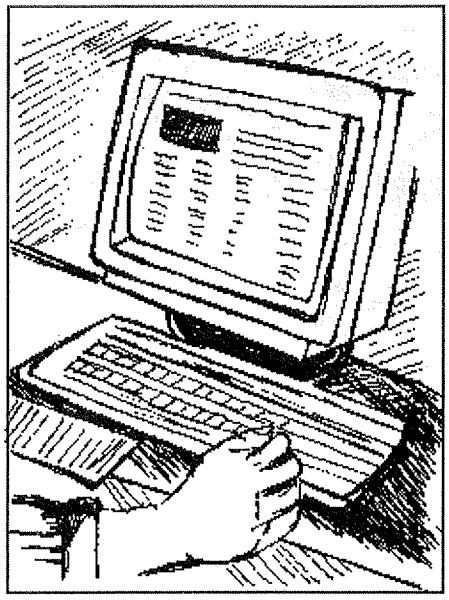
(Ciobanu et.al., Genetics. 2001 Nov; 159(3):1151-62.).

EXPLANATION OF STATISTICS USED IN THIS REPORT

Pigs treated alike vary in performance due to their different genetic makeup and to environmental effects we cannot completely control. When a group of pigs is randomly allotted to treatments it is nearly impossible to get an "equal" group of pigs on each treatment. The natural variability among pigs and the number of pigs per treatment determine the expected variation among treatment groups due to random sampling.

At the end of an experiment, the experimenter must decide whether observed treatment differences are due to "real" effects of the treatments or to random differences due to the sample of pigs assigned to each treatment

Statistics are a tool used to aid in this decision. They are used to calculate the probability that observed differences between treatments were caused by the luck of the draw when pigs were assigned to treatments. The lower this probability, the greater confidence we have that "real" treatment effects exist. In fact when this probability is less than .05 (denoted **P < .05** in the articles), there is less than a 5% chance (less than 1 in 20) that observed treatment differences were due to random sampling. The conclusion then is that the treatment effects are "real" and caused different performance for pigs on each treatment. But bear in mind that if the experimenter obtained this result in each of 100 experiments, 5 differences would be declared to be "real" when they were really due to chance. Sometimes the probability value calculated from a statistical analysis is P < .01. Now the chance



that random sampling of pigs caused observed treatment differences is less than 1 in 100. Evidence for real treatment differences is very strong.

It is commonplace to say differences are significant when P <.05, and highly significant when P < .01. However, P values can range anywhere between 0 and 1. Some researchers say that there is a tendency that real treatment differences exist when the value of P is between .05 and .10. Tendency is used because we are not as confident that differences are real. The chance that random sampling caused the observed differences is between 1 in 10 and 1 in 20.

Sometimes researchers report **standard errors of means (SEM)** or **standard errors (SE)**. These are calculated from the measure of variability and the number of pigs in the treatment. A treatment mean may be given as 11 □ .8. The 11 is the mean and the .8 is the SEM. The SEM or SE is added and subtracted from the treatment mean to give a range. If the same treatments were applied to an unlimited number of animals the probability is .68 (1 = complete certainty) that their mean would be in this range. In the example, the range is 10.2 to 11.8.

Some researchers report **linear (L)** and **quadratic (Q)** responses to treatments. These effects are tested when the experimenter used increasing increments of a factor as treatments. Examples are increasing amounts of dietary lysine or energy, or increasing ages or weights when measurements are made. The L and Q terms describe the shape of a line drawn to describe treatment means. A straight line is linear and a curved line is quadratic. For example, if finishing pigs were fed diets containing .6, .7, and .8% lysine and gained 1.6, 1.8 and 2.0 lb/day, respectively we would describe the response to lysine as linear. In contrast, if the daily gains were 1.6, 1.8, and 1.8 lb/day, the response to increasing dietary lysine would be quadratic. Probabilities for tests of these effects have the same interpretation as described above. Probabilities always measure the chance that random sampling caused the observed response. Therefore, if P < .01 for the Q effect was found, there is less than a 1% chance that random differences between pigs on the treatments caused the observed response.

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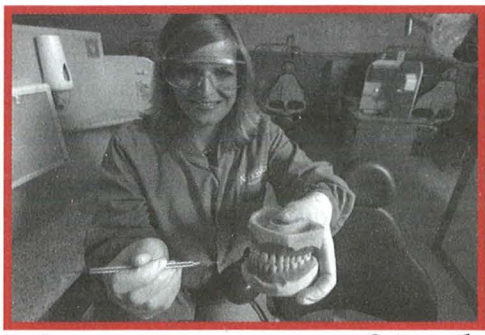

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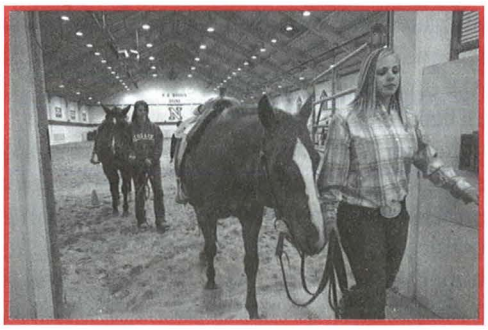

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